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EDITOR: **James W. Berry**, Department of Biological Sciences, Butler University, Indianapolis, Indiana 46208. Telephone: (317)-940-9344; FAX: (317)-940-9519; e-mail: jwberry@butler.edu

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Cover photo: Photo of a cluster of hibernating bats, the Indiana myotis, *Myotis sodalis*. Indiana myotis hibernate in large numbers in tight clusters in a very few caves, with about 90% of the Indiana myotis that hibernate in five caves. There are an estimated 36,000 bats in this cluster in a cave in Crawford County, Indiana. Indiana myotis are listed as endangered, but their range-wide population has continued to decrease. Photo by John O. Whitaker, Jr. (See related article on page 61).

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STATHMIN BINDS HEAT-SHOCK-PROTEIN 70

C.S. King, W. Ko, J. Aloor, D.A. Prentice and **J.P. Hughes**: Indiana State University, Terre Haute, Indiana 47809 USA

ABSTRACT. Stathmin is a cytosolic protein postulated to act as an intracellular relay in cell signaling. It interacts via coiled-coil motifs with several proteins, including BiP/Grp78 and Hsc70, two members of the heat-shock-70 (Hsp70) family of proteins. In the current study, a glutathione-S-transferase (GST)-stathmin fusion protein was used to determine whether stathmin also binds to Hsp70, a third member of the Hsp70 family. Stathmin-binding proteins were examined in Nb2 cells, a rat lymphoma, and in PC12 cells, a rat pheochromocytoma. Prolactin (PRL)-stimulated Nb2 cells were used as the primary source of cytosol and membrane proteins because it has been demonstrated that PRL induces expression of Hsp70 in these cells. PC12 cells were used because they are of neuronal origin, and stathmin appears to play important roles in neural signaling. The results showed that stathmin binds multiple Nb2- and PC12-cell proteins. Immunostaining confirmed that stathmin binds to tubulin and showed for the first time that stathmin binds to Hsp70.

Keywords: Stathmin, Nb2, PC12, Hsp70, glutathione-S-transferase

Stathmin is a ubiquitous, cytosolic phosphoprotein believed to be involved in intracellular signaling (Sobel 1991). Though its signaling mechanism is yet unclear, stathmin is known to associate with several proteins via coiled-coil interactions (Larsson et al. 1999; Li & Cohen 1996; Maucuer et al. 1995; Redeker et al. 2000). Proteins found to interact with stathmin include tubulin (Belmont & Mitchison 1996; Curmi et al. 1997; Gradin et al. 1998; Horwitz et al. 1997; Jourdain et al. 1997; Larsson et al. 1999; Redeker et al. 2000), the tumor susceptibility protein TSG101 (Li & Cohen 1996; Maucuer et al. 1995), the serine/threonine kinase KIS (Maucuer et al. 1995), and members of the heat-shock-70 (Hsp70) family of proteins (Manceau et al. 1999; Maucuer et al. 1995). The two members of the Hsp 70 family shown to bind to stathmin are BiP/GRP 78 (Maucuer et al. 1995) and Hsc70 (Manceau et al. 1999). Evidence suggests that Hsc70 preferentially binds to unphosphorylated stathmin (Manceau et al. 1999), as does tubulin (Curmi et al. 1997; Gradin et al. 1998; Horwitz et al. 1997; Jourdain et al. 1997).

Interaction of unphosphorylated stathmin with tubulin appears to increase the catastrophe rate of microtubules (Belmont & Mitchison 1996). Stathmin may promote shortening of microtubules by sequestering tubulin or by directly interfering with microtubule polymerization. The importance of the interaction of stathmin with members of the Hsp family is less well understood. However, interaction of stathmin with members of the Hsp70 family opens additional pathways for regulation of cell growth and development by stathmin.

Given the known interactions with other members of the Hsp70 family, we attempted to determine whether Hsp70 itself interacts with stathmin using a probe consisting of stathmin coupled to glutathione-S-transferase (GST). The probe also was used to identify stathmin-binding proteins in the Nb2 lymphoma and PC12 cells.

METHODS

Ovine prolactin (oPRL; NIH P-1-2) was obtained from the Hormone Distribution Program, NIH (Bethesda, Maryland). Anti-Hsp70 (K-20), which does not cross-react with Hsc70, was purchased from Santa Cruz Biotechnology, Inc. (Santa Cruz, California). Anti-tubulin and anti-glutathione-S-transferase were obtained from Sigma Chemical Company (Saint Louis, Missouri).

GST-stathmin fusion protein.—A cDNA coding for the entire human stathmin gene was cloned into a SmaI/SalI site in the pGEX-5X-1 GST fusion vector (Amersham Pharmacia Biotech, Piscataway, New Jersey). The 5′ end of the construct was sequenced to ensure that the cDNA was cloned into the cor-

rect frame. The vector was transformed into competent *E. coli* TG1 cells and expression of the GST-stathmin fusion protein was induced by addition of isopropyl-β-D-thiogalactoside (IPTG, 100 mM). The stathmin-GST fusion protein was extracted from the TG1 cells according to the manufacturer's recommendations (GST Gene Fusion System, Amersham Pharmacia Biotech), recovered on glutathione-Sepharose, washed, and eluted using glutathione (10 mM). GST was obtained by expression from pGEX-5X-1 not containing an insert. Protein recovered from glutathione-Sepharose was further purified by FPLC on Superdex 75 (Amersham Pharmacia Biotech). Column fractions containing GST-stathmin or GST were collected, re-purified on glutathione-Sepharose, and washed and concentrated in a Centricon 10 (Amicon Inc., Beverly, Massachusetts).

^{35}S-labeling of Nb2 and PC12 proteins.—Nb2 cells, a rat lymphoma, were grown in Fischer's Medium (for leukemic mouse cells) containing 10% horse serum, 10% fetal bovine serum (FBS), 0.1 mM 2-mercaptoethanol, 100 units of penicillin/ml, and 100 μg of streptomycin/ml (Meyer et al. 1992). Proliferating Nb2 cells (approximately 2×10^7) were washed and resuspended in 3 ml of a methionine- and cysteine-free medium (Minimum Essential Medium Eagle without methionine, cysteine, Sigma Chemical Company) supplemented with prolactin (PRL, 10 ng/ml), 0.1 mM 2-mercaptoethanol, 10% horse serum and a mixture containing ^{35}S-methionine and ^{35}S-cysteine (1 mCi, Express^{35}S^{35}S, NEN Life Science Products, Boston, Massachusetts). PRL stimulates growth of Nb2 cells in the absence of FBS; thus, FBS could be eliminated from the labeling medium. Reduction of serum promotes stronger labeling of proteins with the ^{35}S-methionine/^{35}S-cysteine mixture, because FBS contains significant amounts of unlabeled cysteine and methionine. PC12 cells were grown in RPMI 1640 supplemented with 10% horse serum, 5% FBS, 100 units of penicillin/ml, and 100 μg of streptomycin/ml. Proliferating PC12 cells (approximately 1×10^7) were suspended in 3 ml of the methionine- and cysteine-free medium supplemented with 10% horse serum, 5% FBS, and 1 mCi of the ^{35}S-methionine/^{35}S-cysteine mixture. FBS was added to the labeling incubation to maintain the viability of the PC12 cells. All cells were incubated in their respective media for 6 h at 37° C in CO_2/air (1:19).

Cells were collected by centrifugation at 400 × g for 4 min at 4° C, washed twice in Tris-saline buffer (50 mM Tris-HCl, 150 mM NaCl, pH 8.0), and resuspended in Tris-saline containing proteinase inhibitors [pepstatin (10 μg/ml), leupeptin (10 μg/ml), and aprotinin (0.2 TIU/ml)]. Nb2 cells were passed 10 times through a 23-gauge needle and centrifuged at 12,000 × g for 1 min at 4° C. The supernatant fraction (cytosol) was removed from the pellet, passed through a 0.45 μm filter, and washed and concentrated in a Centricon 10 (Amicon, Inc.). The crude membrane pellet was suspended in Tris-saline containing 1% Triton X-100, 2mM EDTA and proteinase inhibitors, passed 10 times through a 23-gauge needle, and centrifuged at 12,000 × g. The supernatant fraction containing solubilized membranes was removed for use in binding studies.

Because many of the PC12 cells adhere to the culture dish, they were solubilized at 4° C in Tris-saline buffer containing 1% Triton X-100 and proteinase inhibitors. The solubilized cells were centrifuged at 12,000 × g for 1 min and the supernatant fraction was passed through a 0.45 μm filter. PC12 proteins were washed and concentrated in a Centricon 10.

Binding.—Proteins extracted from PC12 cells (whole cell) and from Nb2 cells (cytosol or crude membrane) were incubated with 20 nanomoles of the GST-stathmin fusion protein or equivalent amount of GST protein for approximately 18 h at 25° C. Incubations were performed in a Tris-saline buffer (50 mM Tris-HCl, 150 mM NaCl) containing the proteinase inhibitors. The pH of the buffer was adjusted to 8.0 or 6.8 as noted in figure legends, Triton X-100 was maintained at 0.05–0.1% in solutions containing solubilized membranes, and $MgCl_2$ was added to a final concentration of 5 mM where noted. After incubation, 25 μl of packed glutathione-Sepharose 4B was added to each reaction and the tubes were shaken gently for 30 min at 25° C. The amount of glutathione on the Sepharose was sufficient to bind all the GST (free or associated with the stathmin-GST fusion protein) in the reaction. Therefore, the beads rapidly precipitated the stathmin-GST fusion protein and proteins associated with the fusion protein. The Sepharose beads were collected by centrifugation,

washed 3 times in Tris-saline buffer, and stathmin-GST fusion protein were separated from the Sepharose (solubilized) using 10 mM glutathione.

Electrophoresis and immunostaining.—In each experiment, equal volumes of the glutathione eluates were analyzed by sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) as described by Laemmli (1970) on 10% polyacrylamide gels. Electrophoresis was run on a Mini-Protean II vertical gel system at a constant voltage of 125 V for approximately 90 min, at which time the dye front reached the bottom of the gel. Proteins in the gels were electrophoretically transferred (60 volts, 1 h) to nitrocellulose paper (0.22 μm) using a Trans-Blot Cell (Bio-Rad Laboratories, Hercules, California) and the buffer system described by Towbin et al. (1979). Open sites on the nitrocellulose paper were blocked by incubation in binding buffer (100 mM Tris-HCl, 0.15 M NaCl, 0.1% BSA, pH 8.0) containing 5% non-fat dry milk. After three 10-min washes in binding buffer, the nitrocellulose was incubated for 18–20 h in binding buffer containing the primary antibody. Unbound antibody was removed by three 10-min washes in binding buffer, and the nitrocellulose was incubated for 1 h with a secondary antibody coupled to alkaline phosphatase. Unbound antibody was removed by three 10-min washes in binding buffer, and the alkaline phosphatase was detected using OneStep BCIP/NBT (Pierce Chemical Co., Rockford, Illinois).

RESULTS

The GST-stathmin fusion protein and GST expressed at high levels in the gene fusion system and provided relatively pure probes for analyzing stathmin-binding proteins.

The GST-stathmin fusion protein bound several Nb2-cell proteins. Figure 1 shows ^{35}S-labeled proteins isolated on glutathione-Sepharose from cytosol and crude-membrane extracts after addition of no probe (C), GST (T) or GST-stathmin (S) at pH 8.0. Several cytosolic proteins (noted by asterisks on figure) in the 40-92-kDa range were more prominent in the GST-stathmin lane, and thus may represent protein bound to stathmin directly or via another stathmin-binding protein. A prominent 50 kDa-protein preferentially bound by GST-stathmin appeared to be present in both the cytosol and membrane fractions. A protein(s) migrating at approximately 70 kDa appeared to be represented in both cytosol and membrane fractions, but was more prominent in the membrane fraction.

It has been shown (Curmi et al. 1997) that the stathmin-tubulin interaction is stronger at a pH below 7.0 and in the presence of $MgCl_2$. Therefore, binding of stathmin to ^{35}S-labeled proteins extracted from Nb2 cells was examined at pH 6.8 in the presence of 5 mM $MgCl_2$ (Fig. 2). The pattern of proteins bound by the GST-stathmin probe clearly differed from that obtained at pH 8.0. Several proteins in the 25-69-kDa range (noted by asterisks on figure) were present at pH 6.8. A strong protein doublet migrating at approximately 60 kDa was recognized by the GST-stathmin probe in both the cytosol and membrane fractions at pH 6.8. There was no comparable doublet at pH 8.0, though a protein of approximately 60 kDa was identified in experiments performed at pH 8.0.

The GST-stathmin probe also recognized multiple proteins in the 46-92-kDa range (noted by asterisks on figure) in PC12 cells (Fig. 3). A couple of the proteins migrated with apparent molecular weights similar to those of proteins identified in Nb2 cells, but it is unknown whether the proteins identified in the two cells are related. In PC12 cells, a strong protein doublet migrating with an apparent molecular weight of less than 30 kDa was present in all lanes. This doublet was recognized by anti-transferase antibody (data not shown) suggesting that the proteins represent endogenous GST that binds directly to the glutathione-Sepharose beads.

To determine whether stathmin binds Hsp70, ^{35}S-labeled proteins extracted from Nb2 cells at pH 8.0 were probed with anti-Hsp70 antiserum. The antiserum, which recognizes Hsp70 but not Hsc70, identified a protein at 70-kDa (noted by asterisk on figure) among the proteins recognized by the GST-stathmin fusion protein (Fig. 4A). The immunostained protein appeared to match a ^{35}S-labeled protein (noted by asterisk on figure) isolated using the GST-stathmin probe (Fig. 4B). Thus, the data suggest that stathmin associates with Hsp70 in Nb2 cells.

Stathmin also appeared to bind tubulin-like proteins from Nb2 cells. When binding was performed at pH 6.8, the GST-stathmin fraction contained two proteins recognized by an

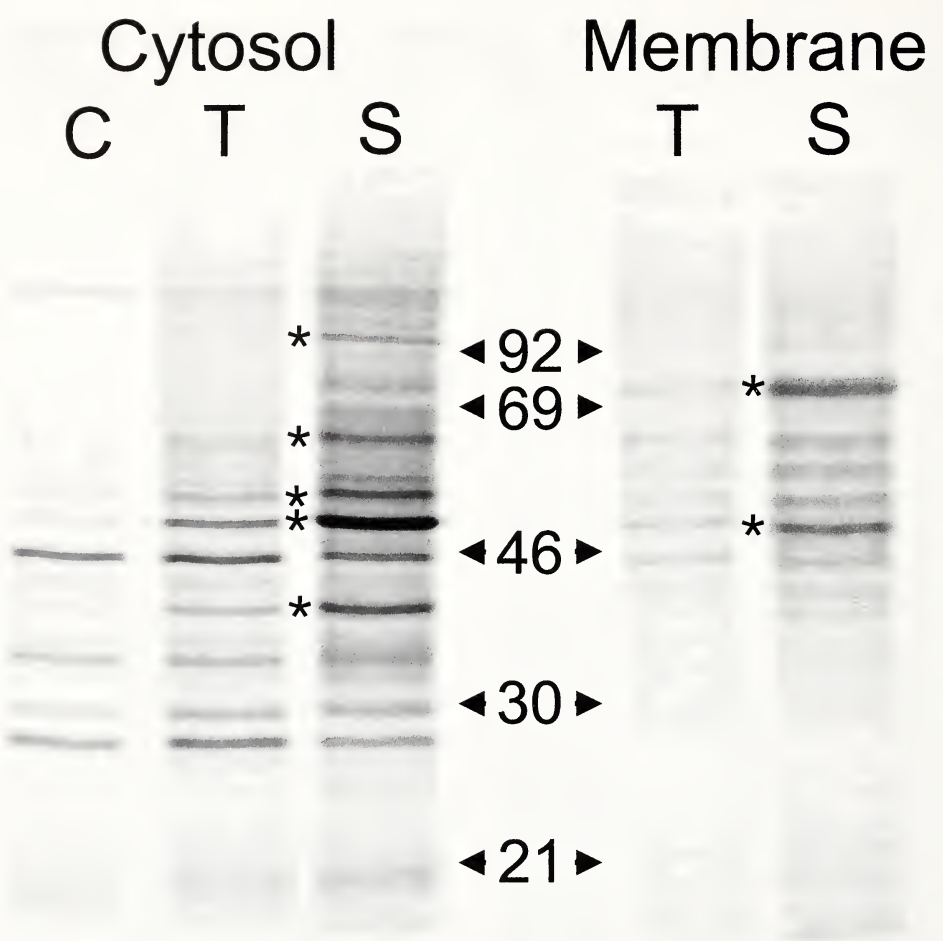

Figure 1.—^{35}S-labeled proteins in cytosol and membrane fractions from Nb2 cells incubated at pH 8.0 with no additions (C), GST (T) or GST-stathmin (S). Proteins were isolated on glutathione-Sepharose and analyzed by SDS-PAGE and autoradiography. Numbers in center show the positions of molecular-weight standards. Asterisks indicate bands that are more prominent in the GST-stathmin lane.

anti-tubulin antiserum (Fig. 5). The two proteins migrated with apparent molecular weights of 55 kDa (Fig. 5), which are consistent with the molecular weights of α and β tubulin. The 55-kDa proteins were not present in control lanes. Tubulin-like proteins also were not identified when binding was performed at pH 8.0 instead of 6.8, consistent with earlier results (Curmi et al. 1997) showing that stathmin binds tubulin better at the lower pH.

DISCUSSION

Stathmin is believed to interact with several cellular proteins, including two member of the Hsp70 family (Manceau et al. 1999; Maucuer et al. 1995). Although only two Hsp70-related proteins, BiP/Grp78 (Maucuer et al. 1995) and Hsc70 (Manceau et al. 1999), have been identified as stathmin-binding proteins, it seems possible that other members of the Hsp70 family may also associate with stathmin. Moreover, in view of its postulated role as an intracellular relay protein (Sobel 1991) and its broad distribution, stathmin likely associates with additional proteins. In the current study we have examined whether stathmin binds to additional proteins, including additional members of the Hsp70 family.

Stathmin appears to bind Hsp70. The stathmin-GST fusion protein isolated a protein that reacts with an antibody specific for Hsp70.

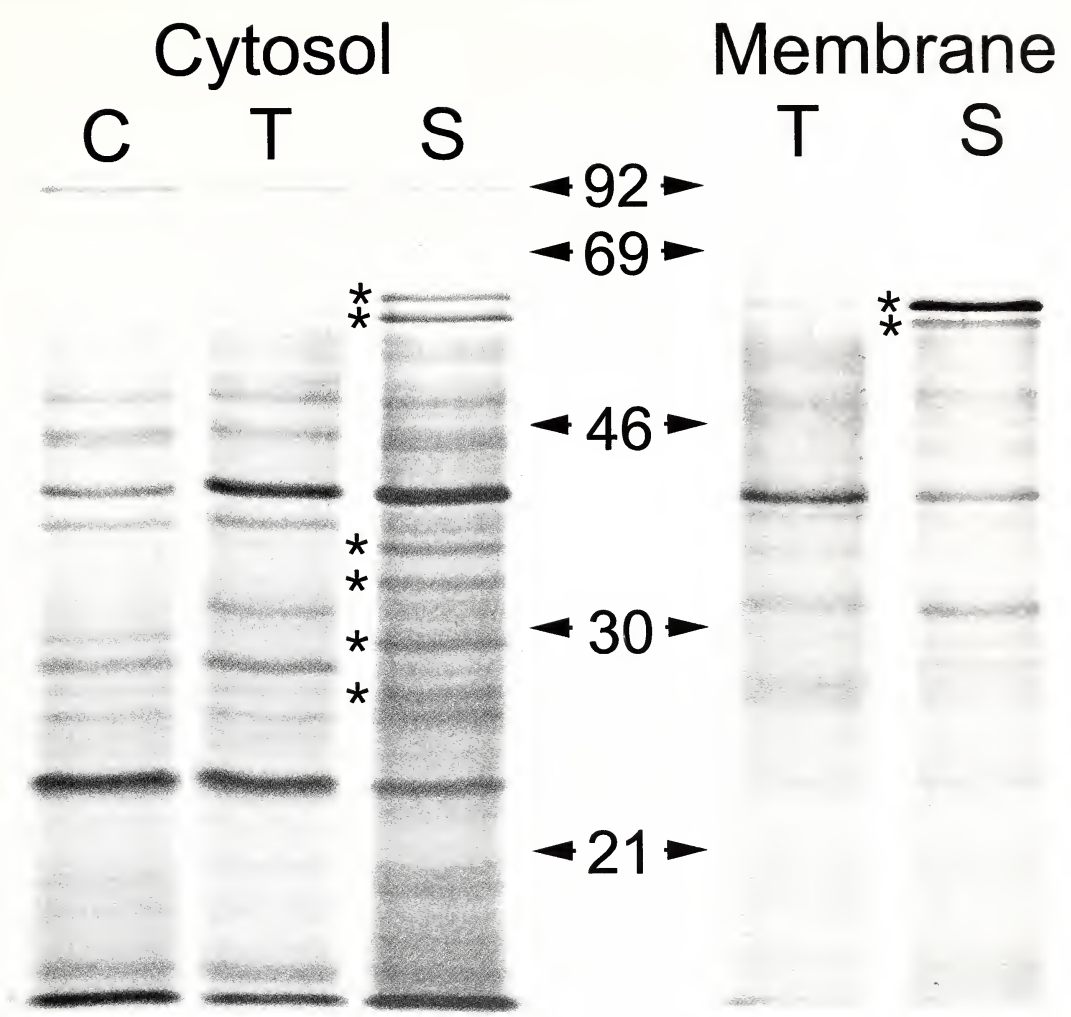

Figure 2.—^{35}S-labeled proteins in cytosol and membrane fractions from Nb2 cells incubated at pH 6.8 and 5 mM $MgCl_2$ with no additions (C), GST (T) or GST-stathmin (S). Proteins were isolated on glutathione-Sepharose and analyzed by SDS-PAGE and autoradiography. Numbers in center show the positions of molecular-weight standards. Asterisks indicate bands that are more prominent in the GST-stathmin lane.

The immunostained protein is unlikely to be Hsc70 because studies by the vendor indicate antibody used in the study does not cross-react with Hsc70. In Nb2 cells, expression of Hsp70 is induced by PRL and other mitogens (de Toledo et al. 1987; Horn et al. 1994); thus, Hsp70 levels would be expected to be elevated in the asynchronous, PRL-treated cells used in the studies. Induction of Hsp70 appears to be closely associated with Nb2-cell proliferation, but the exact role of Hsp70 is unclear. It is interesting that PRL and phorbol esters induce Hsp70 and stimulate phosphorylation of stathmin in Nb2 cells because Manceau et al. (1999) have suggested that phosphorylated forms of stathmin have reduced affinity for Hsp70-like proteins. Perhaps stathmin sequesters Hsp70, and stathmin phosphorylation releases Hsp70 so it can participate in mitogenesis or the response to heat shock. This type of regulation would be consistent with the stathmin-tubulin interaction where phosphorylation of stathmin decreases its ability to sequester tubulin (Curmi et al. 1997; Gradin et al. 1998; Horwitz et al. 1997; Jourdain et al. 1997). If unphosphorylated stathmin does sequester Hsp70, then phosphorylation of stathmin might be an important

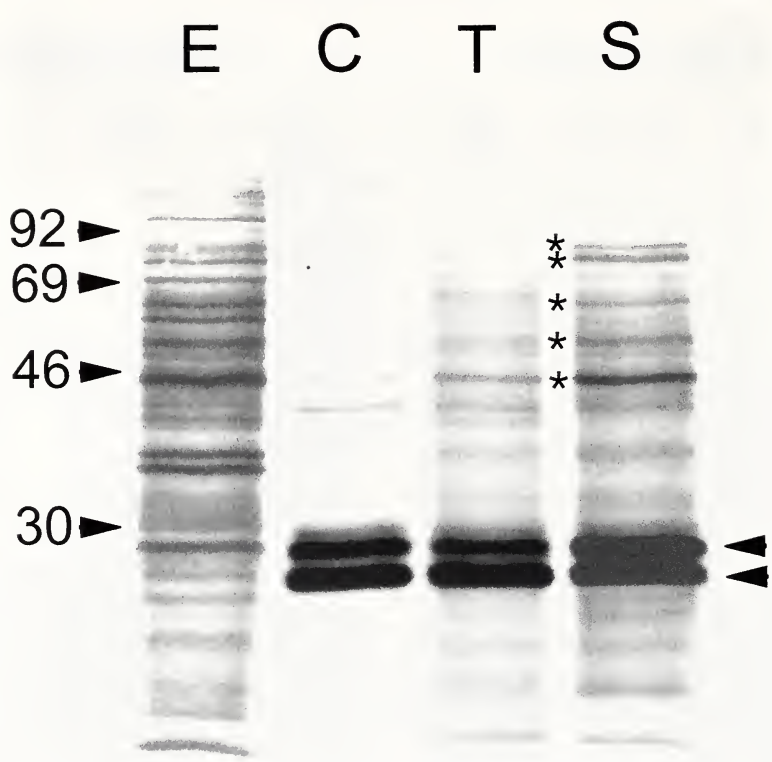

Figure 3.—^{35}S-labeled proteins in whole-cell lysates of PC12 cells (E) were incubated at pH 8.0 with no additions (C), GST (T) or GST-stathmin (S). Proteins in C, T and S were isolated on glutathione-Sepharose and analyzed by SDS-PAGE and autoradiography. Arrowheads at right indicated the positions of proteins recognized by anti-GST antibodies. Numbers at left show the positions of molecular-weight standards. Asterisks indicate bands that are more prominent in the GST-stathmin lane.

consequence of heat shock as has been shown in HeLa cells (Beretta et al. 1995). In any case, these data suggest, for the first time, that stathmin binds Hsp70. It is possible, therefore, that stathmin-Hsp70 interaction participates in signaling in Nb2 cells.

Stathmin also associated with ^{35}S-labeled proteins with molecular weights (55 kDa) approximating those of α and β tubulin. A ^{35}S-labeled protein doublet that might represent α and β tubulin was observed when binding to the stathmin fusion protein was performed at pH 6.8, but not when binding was performed at pH 8.0. Similarly, the anti-tubulin antiserum recognized a doublet only when binding to stathmin was performed at pH 6.8 in the presence of $MgCl_2$, consistent with earlier studies (Curmi et al. 1997). Better binding at a lower pH (i.e., 6.8) has not been shown for other stathmin-binding proteins. In fact, experiments in this study suggest that stathmin may preferentially bind to a different set of proteins at higher pH (i.e., 8.0 *vs.* 6.8). The dependence on pH could reflect many factors, including inherent differences among proteins, requirements for cofactors, or protein modifications such as phosphorylation. The weakness of the ^{35}S-labeled bands and the tubulin immunostaining may reflect the low affinity of the stathmin-tubulin interaction (Curmi et al. 1997) combined with the extensive washing employed in these studies.

Several other putative stathmin-binding proteins were isolated using the GST-stathmin fusion protein to probe Nb2- and PC12-cell extracts. Additional studies will be required to establish the identities of these proteins and their relationships to previously identified stathmin-binding proteins. The two prominent PC12 proteins migrating just below 30 kDa probably represent two or more isozymes of GST because the proteins were isolated using glutathione-Sepharose and recognized by an anti-GST antibody. The PC12 GST, which

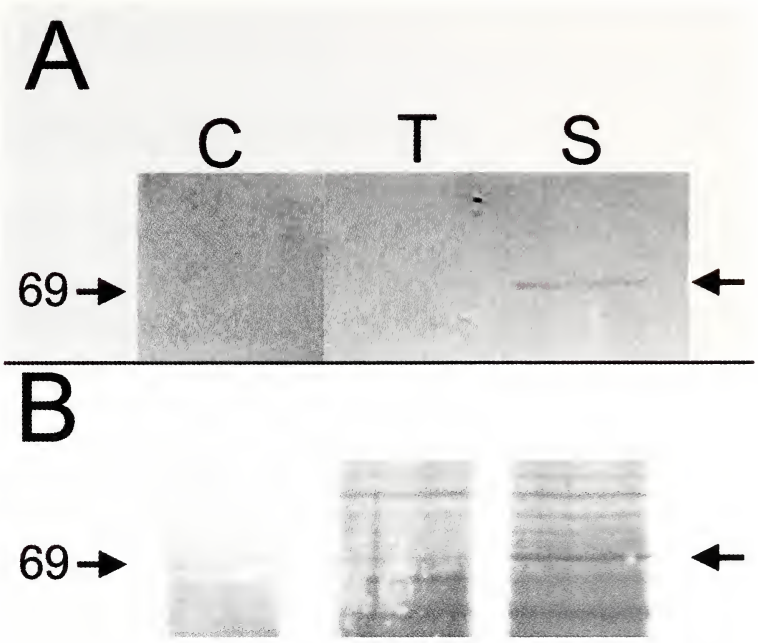

Figure 4.—^{35}S-labeled proteins in a cytosol fraction from Nb2 cells incubated at pH 8.0 with no additions (C), GST (T) or GST-stathmin (S). Proteins were isolated on glutathione-Sepharose and analyzed by SDS-PAGE. Proteins were transferred to nitrocellulose and probed with anti-Hsp70 (A) or analyzed by autoradiography (B). In Fig. 4A, the arrow at right shows the position of a 70-kDa band recognized by the anti-Hsp70 antibody. In Fig. 4B, the arrow at right shows the position of a band that is more prominent in the GST-stathmin lane. The number at left shows the positions a 69-kDa standard.

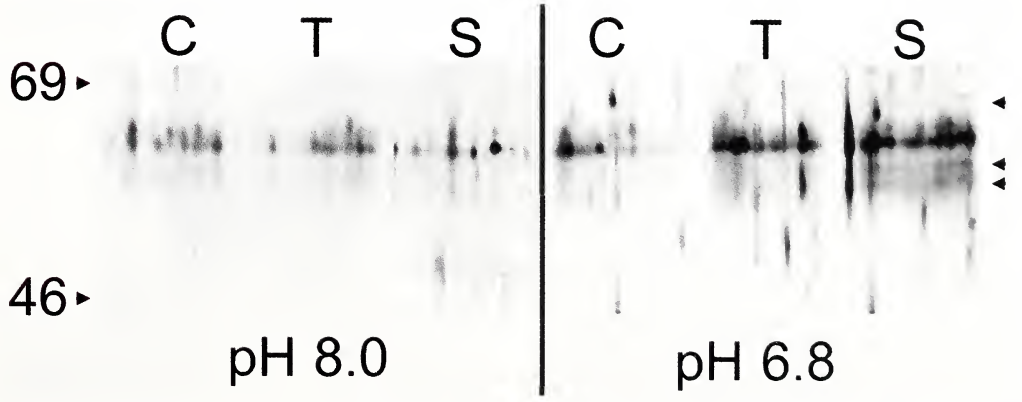

Figure 5.—Proteins in cytosol and membrane fractions from Nb2 cells incubated at pH 8.0 or pH 6.8 and 5 mM $MgCl_2$ with no additions (C), GST (T) or GST-stathmin (S). Proteins were isolated on glutathione-Sepharose, separated by SDS-PAGE, transferred to nitrocellulose and probed with anti-tubulin. Arrowheads at right show the positions of proteins recognized by the anti-tubulin antibody, including a doublet precipitated by the GST-stathmin probe at pH 6.8. The numbers at left show the positions of molecular-weight standards.

may have been induced by mitogens in the serum (Nur-E-Kamal et al. 2000), approximated the amount of GST/GST-stathmin used to probe the extracts.

Overall, the results show that stathmin binds Hsp70 as well as BiP/Grp78 (Maucuer et al. 1995) and Hsc70 (Manceau et al. 1999). Thus, some of the growth-promoting and developmental actions linked to stathmin may involve interaction with Hsp70. Furthermore, the GST-stathmin fusion protein, which proved to be a good probe for stathmin-binding proteins, identified additional potential binding proteins. It seems likely that stathmin associates with multiple binding partners; consistent with the proposal that stathmin acts as an integrative, relay protein (Sobel 1991).

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TWENTY-FIVE YEARS OF FOREST SUCCESSION IN HAPPY VALLEY, JEFFERSON COUNTY, INDIANA

Lindsay Betz and **Paul C. MacMillan:** Department of Biology, Hanover College, Hanover, Indiana 47243 USA

ABSTRACT. In April 1974 a tornado severely damaged the sugar maple/Ohio buckeye forest in Happy Valley, Jefferson County, Indiana. Virtually all canopy trees were destroyed or damaged. In the fall of 1974, the forest was sampled on three transects across the valley. The most damaged portion of the valley forest was characterized as a sugar maple/white oak spp./white ash community. Seven years later the forest was sampled on the same transects, and the most damaged part was described as a sugar maple/elm spp./ Ohio buckeye/redbud community. Fifteen years after the tornado the forest was sampled on the same transects, and the most damaged part was described as a sugar maple/slippery elm community. Twenty and twenty-five years after the tornado the most damaged portion of the forest is still a sugar maple/ slippery elm community, with chinkapin oak, Ohio buckeye, American basswood, white ash and hackberry of secondary importance. The less damaged portion of the valley is dominated by sugar maple, with black walnut, box-elder, white ash, slippery elm and red oak of secondary importance. Important changes in overall forest structure that occurred between 20 and 25 years after the tornado are described, and predictions of future changes are included.

Keywords: Forest composition, forest structure, Jefferson County, Indiana, oaks, Ohio buckeye, secondary succession, slippery elm, sugar maple

Temperate forests in the midwestern United States have been subjected to natural disturbances since they evolved. One dramatic cause of such disturbance is tornadic winds. On the afternoon of 3 April 1974, a series of tornadoes ripped across Indiana. One traveled through the town of Hanover, traversed the Hanover College campus, and continued northeast across the adjacent wooded Happy Valley creating extensive damage. The tornado left nine people dead in the county and damaged nearly every building on the college campus (Larking 1974; Anonymous 1974).

Bailey and MacMillan (1977) completed a census of the Happy Valley forest in the fall of 1974 and found that 90% of the canopy vegetation was removed and approximately one-third of the trees (dbh $\geq$ 5 cm) had been destroyed. The valley floor was so littered with logs that one was forced to climb over them. Forest destruction was most severe at the northern end of the Valley. Destruction of the canopy allowed sunlight to reach the forest floor, which in turn supported abundant herbaceous plant growth. Within a few years, woody shrubs and tree saplings emerged above the piles of fallen stems and branches; and many of the damaged standing stems branched profusely and leafed out. After seven years (1981), emergent trees began to form a canopy creating an environment that supported more shade tolerant plants (Martin & MacMillan 1982).

Data on the Happy Valley forest, gathered before the tornado of 1974, was not systematically collected; but it suggested that the forest had been a sub-climax, sugar maple/Ohio buckeye community. The summer following the tornado, Bailey & MacMillan (1977) established three west-to-east transects across the valley to census the forest. They found the remaining forest to be composed primarily of sugar maple. White ash, elm spp. and white oak spp. were of secondary importance, while Ohio buckeye was a very minor component of the forest. Prior to and since the tornado, sugar maple is the dominant species. However, many changes have taken place in the sub-dominant or secondary tree species since the tornado. Seven years later (1981), the same three transects were recensused. The dominant species in the most severely dam-

aged (northern) portion of the valley was sugar maple, with Ohio buckeye and slippery elm of secondary importance. In the less disturbed (southern) portion of the valley, sugar maple was by far the most dominant tree with white ash, white oak spp., and box-elder of secondary importance (Martin & MacMillan 1982). In 1989, 15 years after the tornado, the valley forest was described as a sugar maple/slippery elm community (MacMillan 1996). American basswood, black maple and sycamore were of secondary importance in the more damaged (northern) part of the valley, while black walnut, white ash, northern red oak, and box-elder were of secondary importance in the less damaged (southern) portion of the valley.

The objectives of this present study were to (1) describe the composition and structure of the Happy Valley forest 20 and 25 years after the tornado of 1974, (2) compare the current valley forest composition and structure with the predictions made 15 years after the tornado, and (3) predict future changes in the forest composition and structure.

STUDY SITE

Happy Valley is located at 38°43′N and 85°27′W, just east of the Hanover College campus, in Jefferson County, Indiana. The valley contains a small intermittent stream that drains from the northwest to the southeast and empties into the Ohio River. The east- and west-facing sides descend steeply (25–50% slope) *ca.* 250 ft (75 m) to a narrow valley floor. The wooded valley slopes are composed of Eden-Caneyville soils, and the valley floor is composed of Dearborn channery silt loam soils (Nickell 1985). Three west-to-east transects were laid out in 1974. These transects have been used in each subsequent survey (Fig. 1). Transects 1 and 2 are located in the more severely damaged north and central areas of the valley, Transect 3 is in the less-damaged southern portion.

METHODS

The point-center-quarter method described by Cottam & Curtis (1956) was used to sample the trees at 15 m intervals along each of three transects. Transect 1 (northern) is 345 m long and included 92 trees (23 points); Transect 2 (middle) is 405 m long and included 109 trees (27 points); Transect 3 (southern) is 390 m long and included 104 trees (26 points).

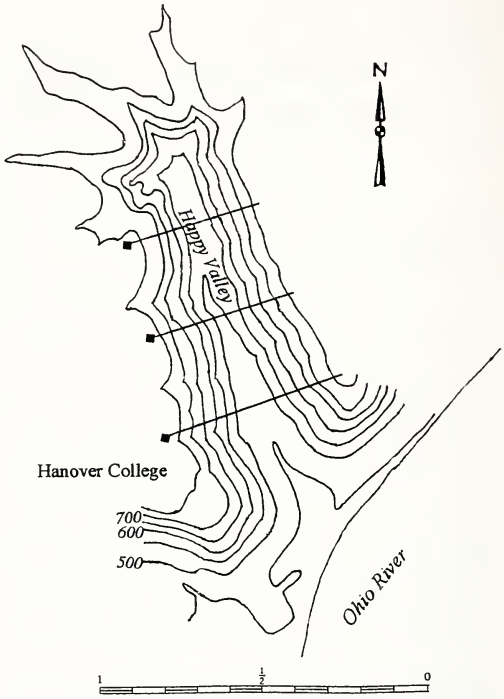

Figure 1.—Map of Happy Valley, Jefferson County, Indiana, showing the location of the three transects described in the text. Transect 1 is at the top, Transect 3 is at the bottom. Contour interval is 50 feet (~ 15 meters).

Data collected in the field included diameter at breast height (dbh) in cm, point-to-tree distance in meters, tree species, and any notes that might prove to be important such as the death of a previously measured tree along the transect line. Relative density, relative frequency, and relative dominance were calculated for each species using the methods of Cox (1990), and those values were summed to determine importance values. Finally, comparisons were made using Sorenson's quotient of similarity (Brower et al. 1990) and the Bray & Curtis (1957) similarity index. Sorenson's quotient of similarity was calculated as:

$$CC_S = 2C/(s_1 + s_2)$$

where C is the number of species common to both sites, s_1 is the number of species at site 1, and s_2 is the number of species at site 2. The Bray & Curtis similarity index was calculated as:

$$CC_B = (2W/(a + b))\ 100$$

where W is the sum of the lower importance values for those species which are common to both sites, a is the sum of importance values

Table 1.—Characteristics of the Happy Valley forest on transect 1 (northern portion) twenty years after the tornado.

Species	Relative density	Relative frequency	Relative dominance	Importance value
Sugar maple				
Acer saccharum Marsh	31.25	27.02	29.17	87.44
Slippery elm				
Ulmus rubra Muhl.	29.17	28.37	18.07	75.61
American basswood				
Tilia americana L.	9.38	12.16	14.00	35.54
Redbud				
Cercis canadensis L.	10.42	8.11	4.10	22.63
Sycamore				
Platanus occidentalis L.	1.04	1.35	19.90	22.29
Chinkapin oak				
Quercus muehlenbergii Engelm.	4.16	5.40	2.72	12.28
Ohio buckeye				
Aesculus glabra Willd.	3.12	4.05	1.63	8.80
White ash				
Fraxinus americana L.	3.12	4.05	1.37	8.54
Hackberry				
Celtis occidentalis L.	3.13	2.70	0.75	6.58
Bitternut hickory				
Carya cordiformis (Wangenh.) K. Koch	1.04	1.35	3.82	6.21
Chokecherry				
Prunus virginiana L.	2.08	2.70	0.54	5.32
Royal paulownia				
Paulownia tomentosa (Thumb.) Sieb. & Zucc.	1.04	1.35	2.60	4.99
Northern red oak				
Quercus rubra L.	1.04	1.35	1.44	3.83
			Sum	300.06

for all the species at site 1, and b is the sum of importance values for all the species at site 2.

RESULTS

These results are from the censuses in 1994 and 1999 (20 and 25 years after the tornado, respectively). Complete transect data by diameter classes for the 1994 and 1999 censuses were reported by Betz (unpubl. report). This report is a summary of the transect data collected in 1994 and 1999. Data by diameter classes can be found in Betz's report available in the Hanover College Library.

Composition of the forest after 20 years (1994).—On Transect 1, across the northern portion of the valley, sugar maple and slippery elm were the dominant trees (importance values 87 and 76, respectively) based on their large relative density, frequency, and dominance (i.e., dbh) (Table 1). American basswood, redbud and sycamore were of secondary importance (importance values 36, 23 and 22, respectively). The frequency and diameter of basswood contributed to its importance value. The importance value of redbud was due to its density and frequency. There was one large sycamore, with a badly damaged top, accounting for its importance. Eight other species, including chinkapin oak, Ohio buckeye, white ash, and hackberry, had lower importance values (Table 1).

On Transect 2, across the middle portion of the valley, sugar maple was the dominant tree (importance value 101) and slippery elm ranked second (importance value 64), again due to their large relative density, frequency, and dominance (Table 3). Trees of secondary importance included chinkapin oak, Ohio buckeye, white ash, box-elder, and hackberry (importance values 33, 26, 18, 14, and 11, respectively). The large diameter of a few chinkapin oaks contributed to its importance value. The importance value of Ohio buckeye was due to many small-diameter stems. Nine

Table 2.—Characteristics of the Happy Valley forest on transect 1 (northern portion) twenty-five years after the tornado.

Species	Relative density	Relative frequency	Relative dominance	Importance value
Sugar maple *Acer saccharum* Marsh	34.05	35.00	41.58	110.63
Slippery elm *Ulmus rubra* Muhl.	25.54	23.75	18.86	68.15
American basswood *Tilia americana* L.	10.63	10.00	14.42	35.05
Chinkapin oak *Quercus muehlenbergii* Engelm.	5.32	6.25	7.94	19.51
Redbud *Cercis canadensis* L.	8.51	7.50	2.85	18.86
Ohio buckeye *Aesculus glabra* Willd.	5.32	5.00	3.04	13.36
Hackberry *Celtis occidentalis* L.	3.19	3.75	1.58	8.52
White ash *Fraxinus americana* L.	2.12	2.50	1.17	5.79
Bitternut hickory *Carya cordiformis* (Wangenh.) K. Koch	1.06	1.25	3.35	5.66
Royal paulownia *Paulownia tomentosa* (Thumb.) Sieb. & Zucc.	1.06	1.25	2.66	4.97
Tulip tree, Yellow poplar *Liriodendron tulipifera* L.	1.06	1.25	1.20	3.51
Northern red oak *Quercus rubra* L.	1.06	1.25	1.15	3.46
Chokecherry *Prunus virginiana* L.	1.06	1.25	0.21	2.52
			Sum	299.99

other species, including red oak, black walnut, and American basswood, had lower importance values (Table 3).

On Transect 3, across the southern end of the valley, sugar maple was the dominant tree (importance 105) with large relative density, frequency, and dominance values (Table 5). Of secondary importance were box-elder, black walnut, white ash, and slippery elm (importance values 31, 31, 30, and 28, respectively). The importance of box-elder and white ash was due to their many small-diameter stems. The importance of black walnut and slippery elm was due to a few large-diameter stems. Ten other species, including chinkapin oak, red oak, Ohio buckeye, and pawpaw, had lower importance values (Table 5).

In 1994, twenty years after the tornado, Transects 1 & 2 (the more damaged part of the valley) remained a sugar maple/slippery elm community with chinkapin oak, Ohio buckeye, basswood, and white ash of secondary importance (Table 7).

Composition of the forest after 25 years (1999).—On Transect 1, sugar maple was the dominant species (importance 111) due to large relative density, frequency, and dominance values (Table 2). Slippery elm was second in importance (68) due to fewer large-diameter trees. American basswood, chinkapin oak, redbud, and Ohio buckeye were of secondary importance (importance values 35, 20, 19, and 13, respectively). The importance of basswood was due to rapid growth of many stems. Chinkapin oak importance was due to its relative dominance. The importance of redbud and Ohio buckeye was due to their many small-diameter stems. Seven other species, including hackberry, white ash, and bitternut hickory, had lower importance values (Table 2).

On Transect 2, the importance value of sugar maple (124) was twice that of slippery elm (62) (Table 4). Large relative density, frequen-

Table 3.—Characteristics of the Happy Valley forest on transect 2 (middle portion) twenty years after the tornado.

Species	Relative density	Relative frequency	Relative dominance	Importance value
Sugar maple *Acer saccharum* Marsh	36.69	30.68	33.82	101.19
Slippery elm *Ulmus rubra* Muhl.	23.85	22.74	17.28	63.87
Chinkapin oak *Quercus muehlenbergii* Engelm.	5.51	6.83	20.23	32.57
Ohio buckeye *Aesculus glabra* Willd.	10.09	11.37	4.84	26.30
White ash *Fraxinus americana* L.	5.50	6.82	5.26	17.58
Box-elder *Acer negundo* L.	4.58	5.68	3.71	13.97
Hackberry *Celtis occidentalis* L.	3.67	3.42	4.03	11.12
Northern red oak *Quercus rubra* L.	0.92	1.14	5.52	7.58
Black walnut *Juglans nigra* L.	0.92	1.14	3.75	5.81
American basswood *Tilia americana* L.	1.83	2.27	0.15	4.25
Pawpaw *Asimina triloba* (L.) Dunal	1.83	2.27	0.11	4.21
Eastern hop hornbeam *Ostrya virginiana* (Mill.) K. Koch	0.92	1.14	0.59	2.65
Sycamore *Platanus occidentalis*	0.92	1.14	0.42	2.48
Osage orange *Maclura pomifera* (Raf.) Schneid.	0.92	1.14	0.16	2.22
Black cherry *Prunus serotina* Ehrh.	0.92	1.14	0.09	2.15
White mulberry *Morus alba* L.	0.92	1.14	0.05	2.11
			Sum	300.06

cy, and dominance values provide the high importance value of sugar maple. Slippery elm values were smaller, and there were fewer large-diameter trees. Of secondary importance were Ohio buckeye, chinkapin oak, white ash, and box-elder (importance values 24, 22, 16, and 15, respectively). The importance of Ohio buckeye was due to many small-diameter stems. A few larger-diameter stems of chinkapin oak contributed to its importance. Eight other species, including hackberry, red oak, black walnut, and American basswood, had lower importance values (Table 4).

On Transect 3, the importance value of sugar maple was 98 (Table 6). Of secondary importance were black walnut, box-elder, white ash, slippery elm, and red oak (importance values 34, 28, 26, 24, and 24, respectively). The importance of box-elder and ash was due to the rapid growth of many small stems. The importance of walnut, slippery elm, and red oak was due to a few larger-diameter stems. Ohio buckeye and chinkapin oak had lower importance values on this transect. Seven other species, including black maple, hackberry, and Osage orange, had lower importance values (Table 6).

Tables 7 and 8 contain data combined from the more heavily damaged northern and middle portions of the valley (Transects 1 and 2) collected 20 and 25 years after the tornado, respectively. These data show changes that

Table 4.—Characteristics of the Happy Valley forest on transect 2 (middle portion) twenty-five years after the tornado.

Species	Relative density	Relative frequency	Relative dominance	Importance value
Sugar maple *Acer saccharum* Marsh	43.11	37.78	43.09	123.98
Slippery elm *Ulmus rubra* Muhl.	21.09	22.22	18.51	61.82
Ohio buckeye *Aesculus glabra* Willd.	9.17	10.00	4.40	23.57
Chinkapin oak *Quercus muehlenbergii* Engelm.	4.59	5.55	12.10	22.24
White ash *Fraxinus americana* L.	6.42	6.66	2.52	15.60
Box-elder *Acer negundo* L.	4.59	5.55	4.46	14.60
Hackberry *Celtis occidentalis* L.	2.75	2.22	4.07	9.04
Northern red oak *Quercus rubra* L.	0.92	1.11	5.25	7.28
Black walnut *Juglans nigra* L.	0.92	1.11	3.49	5.52
American basswood *Tilia americana* L.	1.83	2.22	0.25	4.30
Pawpaw *Asimina triloba* (L.) Dunal	1.83	2.22	0.14	4.19
Black cherry *Prunus serotina* Ehrh.	0.92	1.11	1.04	3.07
Sycamore *Platanus occidentalis* L.	0.92	1.11	0.51	2.54
Osage orange *Machura pomifera* (Raf.) Schneid.	0.92	1.11	0.20	2.23
			Sum	299.98

occurred in the tornado-damaged area, and allow comparison with the less-damaged southern portion (Transect 3). Over the 5 years of this study sugar maple, slippery elm, chinkapin oak, Ohio buckeye, American basswood, and white ash keep the same descending order of importance although the importance value of sugar maple increased about 15%. In 1999, twenty-five years after the tornado, the more damaged (northern and middle) part of the valley remained a sugar maple/slippery elm community with chinkapin oak, Ohio buckeye, basswood, and white ash of secondary importance (Table 8). In the less disturbed southern part of the valley, sugar maple was clearly the dominant tree, with black walnut, box-elder, white ash, slippery elm, and red oak of secondary importance (Table 6).

Similarity indices.—Sorenson's quotient of similarity showed a general increase in similarity within transects from 1989–1994 and into 1999 (Table 9). Transects 1 and 2 (across the northern and middle portions of the valley) had relatively low similarities. Transects 1 and 3 (northern and southern) also had low similarities. Transects 2 and 3 (middle and southern) had the highest similarity indices in each of the three survey years. The comparison of Transects 1 + 2 and 3 showed increasing similarity through time.

The Bray & Curtis similarity index also showed a trend of increasing similarity between transects from 1989–1999 (Table 9), except for the comparison of Transects 1 and 3, which decreased. The similarity between Transects 1 and 2 increased over the 10 years. Likewise, the similarity between Transects 2 and 3 increased somewhat. And the comparison between Transects 1 + 2 and 3 showed an increasing trend of similarity.

Table 5.—Characteristics of the Happy Valley forest on transect 3 (southern portion) twenty years after the tornado.

Species	Relative density	Relative frequency	Relative dominance	Importance value
Sugar maple *Acer saccharum* Marsh	34.83	37.23	33.10	105.16
Box-elder *Acer negundo* L.	14.28	9.57	7.35	31.20
Black walnut *Juglans nigra* L.	6.25	7.44	17.02	30.71
White ash *Fraxinus americana* L.	11.61	11.71	6.47	29.79
Slippery elm *Ulmus rubra* Muhl.	8.04	7.44	12.98	28.46
Northern red oak *Quercus rubra* L.	1.79	1.06	8.43	11.28
Chinkapin oak *Quercus muehlenbergii* Engelm.	3.57	4.25	2.49	10.31
Ohio buckeye *Aesculus glabra* Willd.	4.46	5.32	0.44	10.22
Pawpaw *Asimina triloba* (L.) Dunal	5.36	4.26	0.45	10.07
Osage orange *Maclura pomifera* (Raf.) Schneid.	1.78	2.12	5.20	9.10
Hackberry *Celtis occidentalis* L.	2.68	3.19	0.90	6.77
Black maple *Acer nigrum* Michx.	1.79	2.13	2.25	6.17
Redbud *Cercis canadensis* L.	1.79	2.13	0.27	4.19
Sycamore *Platanus occidentalis* L.	0.89	1.06	2.59	4.54
Black cherry *Prunus serotina* Ehrh.	0.89	1.06	0.06	2.01
			Sum	299.98

Changes in forest structure.—On all three transects the mean point-to-tree distance decreased from 1974–1989 and 1994 (15–20 years) then increased in 1999 (Table 10). Likewise, the mean area per tree decreased from 1974–1989 and 1994; then increased in 1999 on all three transects. In the most damaged portion of the valley (Transects 1 & 2), mean area per tree decreased from 23.7 in 1974, to 11.0 in 1981, to 8.5 in 1989, then increased to 10.3 in 1994 and 11.2 in 1999. On all three transects, the density of trees (stems/ha) increased from 1974–1989 and 1994, then decreased in 1999 (Table 10). The more damaged portion of the valley (Transects 1 & 2) increased in density from 425 in 1974 to 1186 in 1989. These three sets of data illustrate the rapid growth of new trees in Happy Valley following the tornado of 1974, even on the less damaged southern portion (Transect 3).

DISCUSSION

Changes in forest composition.—Over the past 25 years, sugar maple has remained the most important tree in the northern and middle portions (Transects 1 & 2) of Happy Valley. It reproduces and grows successfully in shade and in clearings (Elias 1980). It grows rapidly and resists wind damage. Following the census in 1989, MacMillan (1996) suggested that if the relative density and relative frequency of sugar maple continued to decrease, as they had over the first 15 years, then the fast-growing slippery elm would become the most important tree in this area of the valley. Now

Table 6.—Characteristics of the Happy Valley forest on transect 3 (southern portion) twenty-five years after the tornado.

Species	Relative density	Relative frequency	Relative dominance	Importance value
Sugar maple *Acer saccharum* Marsh	35.08	36.08	27.26	98.42
Black walnut *Juglans nigra* L.	7.90	9.28	17.28	34.46
Box-elder *Acer negundo* L.	11.40	9.27	7.34	28.01
White ash *Fraxinus americana* L.	10.52	10.30	4.68	25.50
Slippery elm *Ulmus rubra* L.	7.01	6.18	10.76	23.95
Northern red oak *Quercus rubra* L.	3.51	3.09	17.08	23.68
Ohio buckeye *Aesculus glabra* Willd.	5.26	6.19	0.47	11.92
Chinkapin oak *Quercus muehlenbergii* Engelm.	4.38	5.15	2.18	11.71
Black maple *Acer nigrum* Michx.	2.63	3.09	4.03	9.75
Hackberry *Celtis occidentalis* L.	4.38	4.12	1.05	9.55
Osage orange *Maclura pomifera* (Raf.) Schneid.	1.76	2.06	5.33	9.15
Pawpaw *Asimina triloba* (L.) Dunal	3.51	2.06	0.34	5.91
Sycamore *Platanus occidentalis* L.	0.88	1.03	2.01	3.92
Redbud *Cercis canadensis* L.	0.88	1.03	0.15	2.06
Black cherry *Prunus serotina* L.	0.88	1.03	0.08	1.99
			Sum	299.98

after an additional 10 years, the importance of sugar maple is increasing: 80 in 1989 (MacMillan 1996), 100 in 1994 (Table 7), and 118 in 1999 (Table 8). Although sugar maple has increased in all three components of the importance value, most of the increase has been in their relative dominance (i.e., basal area). We predict that sugar maple will continue to increase in importance over the next 10 years in this most damaged portion of the valley, then it will decrease as other canopy species increase in density and basal area.

Slippery elm and American elm were both found in this most damaged portion of Happy Valley in 1974 and 1981, although only slippery elm was found in 1986, 1994, and 1999. The American elm apparently died off due to Dutch elm disease, as it had on the adjacent Hanover College campus. Slippery elm grows in many habitats in eastern North America, including dry slopes and well-drained bottomlands. It grows moderately fast and is often found growing with sugar maples (Elias 1980). The rank of slippery elm importance increased from fifth in 1974, to third in 1981, to second in 1989. It has been second to sugar maple since 1989 (Tables 7 & 8). Over the past 25 years the importance value of slippery elm has rested on its relative density and relative frequency, indicative of many smaller stems. It has not become the most important tree in the valley, as was suggested by MacMillan (1996). It now appears that few slippery elm saplings become 5 cm diameter trees (Betz unpubl. data). If this pattern persists, the importance of slippery elm will increase as

Table 7.—Characteristics of the most damaged (northern and middle) portions of the Happy Valley forest twenty years after the tornado.

Species	Relative density	Relative frequency	Relative dominance	Importance value
Sugar maple				
Acer saccharum Marsh	34.64	31.14	33.87	99.65
Slippery elm				
Ulmus rubra Muhl.	26.35	24.55	18.05	68.95
Chinkapin oak				
Quercus muehlenbergii Engelm.	4.87	5.40	11.67	21.94
Ohio buckeye				
Aesculus glabra Willd.	6.83	7.78	3.89	18.50
American basswood				
Tilia americana L.	5.37	6.58	4.91	16.86
White ash				
Fraxinus americana L.	4.39	6.00	4.08	14.47
Redbud				
Cercis canadensis L.	4.88	3.59	1.40	9.87
Hackberry				
Celtis occidentalis L.	3.42	3.00	3.03	9.45
Sycamore				
Platanus occidentalis L.	0.98	1.20	7.12	9.30
Box-elder				
Acer negundo L.	1.96	3.00	1.94	6.90
Northern red oak				
Quercus rubra L.	0.98	1.20	4.29	6.47
Black walnut				
Juglans nigra L.	0.49	0.60	2.59	3.68
Bitternut hickory				
Carya cordiformis (Wangenh.) K. Koch	0.49	0.60	1.31	2.40
Chokecherry				
Prunus virginiana L.	0.98	1.20	0.19	2.37
Pawpaw				
Asimina triloba (L.) Dunal	0.98	1.20	0.08	2.26
Royal paulownia				
Paulownia tomentosa (Thumb.) Seib. & Zucc.	0.49	0.60	0.88	1.97
Eastern hop hornbeam				
Ostrya virginiana (Mill.) K. Koch	0.49	0.60	0.41	1.50
Osage orange				
Maclura pomifera (Raf.) Schneid.	0.49	0.60	0.11	1.20
Black cherry				
Prunus serotina Ehrh.	0.49	0.60	0.07	1.16
White mulberry				
Morus alba L.	0.49	0.60	0.04	1.13
			Sum	300.03

the present stems increase in diameter but over the longer term it will decrease due to lack of recruitment.

The importance of sycamore has changed dramatically over the past 25 years. Sycamore only grows where it has a continual water supply, in this case, only in the valley bottom. Following the tornado of 1974, Bailey (unpubl. data) recorded only three sycamores on Transects 1 & 2, although all were large trees. What did not show in that tree census was the number of large-diameter sycamore logs on the valley floor, making it impossible to walk through the valley. As MacMillan (1996) described, many of the sycamore logs remained rooted and their branches became trees. This is the history of sycamore importance over the past 25 years. In 1974 there were 3 sycamore

Table 8.—Characteristics of the most damaged (northern and middle) portions of the Happy Valley forest twenty-five years after the tornado.

Species	Relative density	Relative frequency	Relative dominance	Importance value
Sugar maple *Acer saccharum* Marsh	38.92	36.47	42.45	117.84
Slippery elm *Ulmus rubra* Muhl.	23.15	22.94	18.62	64.71
Chinkapin oak *Quercus muehlenbergii* Engelm.	4.93	5.88	10.51	21.32
Ohio buckeye *Aesculus glabra* Willd.	7.39	7.65	3.98	19.02
American basswood *Tilia americana* L.	5.91	5.88	5.63	17.42
White ash *Fraxinus americana* L.	4.43	4.70	2.00	11.13
Hackberry *Celtis occidentalis*	2.97	2.95	3.11	9.03
Redbud *Cercis canadensis* L.	3.95	3.53	1.08	8.56
Box-elder *Acer negundo* L.	2.46	2.95	2.76	8.17
Northern red oak *Quercus rubra* L.	0.98	1.18	3.69	5.85
Black walnut *Juglans nigra* L.	0.49	0.59	2.16	3.24
Bitternut hickory *Carya cordiformis* (Wangenh.) K. Koch	0.49	0.59	1.27	2.35
Pawpaw *Asimina triloba* (L.) Dunal	0.99	1.18	0.08	2.25
Royal paulownia *Paulownia tomentosa* (Thumb.) Seib. & Zucc.	0.49	0.59	1.01	2.09
Black cherry *Prunus serotina* Ehrh.	0.49	0.59	0.65	1.73
Tulip tree, Yellow poplar *Liriodendron tulipifera* L.	0.49	0.59	0.46	1.54
Sycamore *Platanus occidentalis* L.	0.49	0.59	0.31	1.39
Osage orange *Maclura pomifera* (Raf.) Schneid.	0.49	0.59	0.12	1.20
Chokecherry *Prunus virginiana* L.	0.49	0.59	0.08	1.16
			Sum	300

stems in the most damaged area, and it ranked second to sugar maple in relative dominance. In 1981 there were 2 stems, and it ranked second to sugar maple. In 1989 there were 4 stems, and it ranked second to sugar maple. In 1994 there were 2 stems, and it ranked fourth to sugar maple. In 1999 there was 1 stem, and it ranked fifteenth in relative dominance. Clearly, the large-diameter stems that remained after the tornado started dropping out after 1989. We suggest that sycamore will remain a very minor component of this damaged portion of the valley for many years to come, until new saplings along the creek are recruited into tree diameter classes. Chinkapin oak is found throughout the American Midwest (Elias 1980). It grows in well-drained soils on upland sites, limestone outcrops, and slopes. Because leaf and bark characters are often not distinct in young oak trees, earlier studies only distinguished "red oak spp." and "white oak spp." In 1989 chinka-

Table 9.—Comparison of Sorenson's quotient of similarity (Brower et al. 1990) and the Bray & Curtis (1957) similarity index between transects from 1989 to 1999.

	1989	1994	1999
Transects 1 & 2			
Sorenson	0.563	0.621	0.593
Bray & Curtis	58.64	66.73	75.79
Transects 1 & 3			
Sorenson	0.600	0.643	0.571
Bray & Curtis	57.30	55.61	55.29
Transects 2 & 3			
Sorenson	0.600	0.839	0.897
Bray & Curtis	61.91	72.20	69.67
Transects 1 + 2 & 3			
Sorenson	0.556	0.800	0.824
Bray & Curtis	60.63	68.04	64.03

pin oak ranked fourth, in 1994 and 1999 it ranked third; each time the importance value was influenced by its large relative dominance value. However, these importance values are 20–30% smaller than those of sugar maple, suggesting that several larger-diameter trees survived the tornado of 1974 because young oak trees would not grow to that diameter in 25 years (Elias 1980). We predict that chinkapin oak will remain an important secondary species in this most damaged portion of the valley and that it will maintain its current rank.

Ohio buckeye is found throughout the Midwestern states in well-drained soils (Elias 1980). It is commonly found as a minor component although the trees are usually large. Its seedlings develop quickly and are very shade-tolerant. Based on unpublished pre-tornado data, Bailey & MacMillan (1977) described

Table 10.—Changes in structure of the Happy Valley forest over 25 years. Pre-tornado conditions (based on unpublished 1973 class data) were: Mean point-to-tree distance = 4.4 m, Mean area/tree = 19.56 m^2, Density of trees = 512/ha. Reference citations (a) Bailey & MacMillan 1977. (b) Martin & MacMillan 1982. (c) MacMillan 1996. (d) Lindsay Betz unpublished.

	1974 (a)	1981 (b)	1989 (c)	1994 (d)	1999 (d)
Transect 1					
Mean point-to-tree distance (m)	5.1	3.1	2.9	2.9	3.2
Mean area/tree (m^2)	25.64	9.60	7.83	8.41	10.24
Density (trees/ha)	391	1041	1180	1231	983
Transect 2					
Mean point-to-tree distance (m)	4.7	3.5	3.0	3.5	3.5
Mean area/tree (m^2)	21.78	12.30	9.21	12.25	12.25
Density (trees/ha)	460	813	1091	864	819
Transect 3					
Mean point-to-tree distance (m)	3.9	3.7	3.4	3.3	3.2
Mean area/tree (m^2)	15.24	13.70	11.57	10.89	13.69
Density (trees/ha)	652	730	857	946	746
Transects 1 & 2					
Mean point-to-tree distance (m)	4.9	3.3	2.9	3.3	3.4
Mean area/tree (m^2)	23.70	11.00	8.51	10.33	11.24
Density (trees/ha)	425	926	1186	1047	901

the middle portion of Happy Valley as a sugar maple/Ohio buckeye sub-climax forest. At that time, Ohio buckeye was a significant component in small- and large-diameter classes, indicating that saplings were being recruited into the "tree" category and that some large trees survived the tornado. In the combined data from Transects 1 and 2, Ohio buckeye ranked a distant second in 1981, ninth in 1989, and fourth in 1994 and 1999. The decrease found in 1989 was due to loss of several large-diameter trees that were damaged in the tornado, managed to survive through 1981, but succumbed by 1984. In 1994 and 1999 the importance of Ohio buckeye was due to many small stems (relative density and frequency). As predicted, the importance value of Ohio buckeye has increased following canopy closure (MacMillan 1996).

American basswood is a fast-growing, long-lived tree common in sugar maple forests (Elias 1980). It grows best in low, moist, well-drained areas, although it grows well on the mesic east-facing slopes of Happy Valley. Following the tornado of 1974, most of the damaged basswood trees coppiced, meaning they sent up several stems from the root crown. In this most damaged portion of the valley, American basswood ranked fourteenth in 1974, seventh in 1981, ninth in 1989, and fifth in 1994 and 1999. These importance values were due to higher relative density and frequency values during the first 20 years. After 25 years, as predicted (MacMillan 1996), the relative dominance (basal area) of basswood is now a significant factor in its importance, indicating the growth of smaller stems into larger diameter classes.

White ash is found across the eastern United States in deep, well-drained soils (Elias 1980). It often grows near streams and along lower slopes of hills and mountains. It grows slowly during the sapling stage and increases in growth rate over the next 50 years. In this most damaged portion of the valley, white ash ranked a distant second in 1974 (based on its relative frequency and dominance), fifteenth in 1981 (based on a very small relative dominance), twelfth in 1989 (with a larger relative dominance), and sixth in 1994 and 1999 based more on relative density and frequency than dominance. These results suggest that a few large white ash trees were damaged but survived the tornado and spread many seeds. The older trees then died, and many saplings were recruited into the smaller diameter classes before the canopy closed, as MacMillan (1996) suggested. There is still a large white ash seedling population in the understory.

Tulip tree (or yellow poplar) is an invader of disturbed sites. It was not found in the most disturbed region of the valley in 1974. Two stems were recorded in 1981, 1 stem in 1989, none in 1994, and 1 stem in 1999. After the 15-year census, MacMillan (1996) predicted that this rapidly growing species would increase in importance over the next 5 years. That has not happened, probably because the canopy closed and few disturbed sites remain.

Changes in forest structure.—Sorenson's quotient of similarity (Brower et al. 1990) suggests that Transects 1 & 2, and Transects 1 & 3 became more similar between 1989 and 1994, and decreased in similarity in 1999 (Table 9). Transects 2 & 3, and 1 + 2 & 3 increased in similarity over the past 10 years. These data suggest that the more damaged northern and central portions of the valley are becoming similar to the less damaged southern portion. The Bray & Curtis (1957) similarity index indicates that Transects 1 & 2 became more similar, Transects 1 & 3 became less similar, and Transects 2 & 3, and 1 + 2 & 3 became slightly more similar over the past 10 years (Table 9). Differences between the two measures of similarity may be due to the factors they compare. Bray and Curtis compare importance values, and Sorenson compares the number of species common to both sites. The trend toward increasing similarity was expected due to the proximity of the sites, the similar soils, seed sources, exposure and climate. This trend of increasing similarity throughout the valley forest is predicted to continue.

The structure of the Happy Valley forest has not continually changed in one direction over the past 25 years (Table 10). Between 15 and 20 years after the tornado (1989–1994) the trend of decreasing values of mean point-to-tree distance and mean area per tree, and the increasing values of tree density (number of trees per hectare), reversed on all three transects. Forest recovery had reached a turning point. After 20 years, the mean point-to-tree distance began to increase suggesting that some trees had died. Likewise, the mean area per tree began to increase suggesting that the

canopy had closed and canopy trees were increasing in diameter. On the more-damaged Transects 1 & 2, the density of trees in 1974 (immediately following the tornado) was lower than on the less damaged Transect 3, as expected due to wind damage. By 1981 (7 years after the tornado) the density of trees on Transects 1 & 2 was higher than on the less-damaged Transect 3, and it remained higher for 25 years. This is explained by the rapid growth of saplings once the canopy had been removed. By 1999 (25 years after the tornado) the density of trees had decreased on all three transects (Table 10). We predict that the mean point-to-tree distance and mean area per tree will continue to increase, and the density of trees will continue to decrease, as the valley forest matures. We attempted to use size structure analysis (Parker 1992; Ziegler 1995) to evaluate the structural dynamics of the most abundant species on each transect. However, our samples were too small, 79% of the stems sampled were less than 25 cm dbh, and historically most of the trees were less than 25 years old due to the tornado of 1974.

In conclusion, we predict that the damaged and undamaged areas of Happy Valley will continue to change in structure and composition and that they will continue to become more similar. Sugar maple will continue to be the dominant tree throughout the valley. If Ohio buckeye continues to increase in importance and establish itself in the canopy, the valley forest may grow back to the same type of community it was before the tornado of 1974.

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DISTRIBUTION RECORDS OF SOUTHERN INDIANA VASCULAR PLANTS

Richard H. Maxwell and **William E. Thomas:** Herbarium, Indiana University Southeast, New Albany, Indiana 47150 USA

ABSTRACT. Collections in the Indiana University Southeast Herbarium were reexamined and entered into a database. New and old collections yielded over 100 new vascular plant county records for Clark, Crawford, Floyd, and Harrison Counties in southern Indiana. Indiana vascular species listed as extirpated (SX), endangered (SE), threatened (ST), or rare (SR) are noted. Others species which may become listed, or are otherwise noteworthy, are briefly discussed.

Keywords: Vascular plants, Indiana, distribution records

The Indiana University Southeast Herbarium serves as an educational and community resource as well as a research exchange facility. The herbarium contains vascular plant collections primarily from Clark, Crawford, Floyd, Harrison, and Jefferson Counties, with occasional collections from Washington, Scott, and Perry Counties. This area of southern Indiana includes the lowermost parts of the following regions: the Mitchell Karst Plain and Knobstone Escarpment Sections of the Highland Rim Natural Region, the Scottsburg Lowland and Muscatatuck Flats and Canyon Sections of the Bluegrass Natural Region, and the Shawnee Hills Natural Region (Homoya et al. 1985). The Indiana University Southeast Herbarium is in the process of creating an online database of its vascular plant collection at www.ius.edu/biology/herbarium/homepage.stm.

Ongoing collecting and review of existing collections in the herbarium have yielded over 100 new vascular plant county records since the publication of the last distribution list from the Indiana University Southeast Herbarium (Maxwell & Emmert 1995). Possible state records and noteworthy collections are listed alphabetically by their scientific names and briefly discussed, followed by a listing of state endangered, threatened, and rare species. Table 1 includes distribution records for Clark, Crawford, Floyd, and Harrison Counties with their collection vouchers. Nomenclature conforms primarily to the *Field Guide to Indiana Wildflowers* (Yatskievych 2000), with authors of plant names, following Brummitt & Powell (1992). Some nomenclature, primarily ferns, grasses, and common names cited, is from Mohlenbrock (1986), Gleason & Cronquist (1991), Homoya (1993) and Yatskievych (2000).

Determination of species status follows the Indiana Natural Heritage Data Center Endangered, Threatened and Rare Vascular Plants list, Indiana, from the Department of Natural Resources (DNR), Nature Preserves (27 January 1998). County status was also determined from the DNR, Nature Preserves web site, www.in.gov/dnr/naturepr (16 November 1999).

Former and present collectors include Indiana University Southeast students, the authors, several volunteers, Ray Weatherholt and several of his biology students from Floyd Central High School.

METHODS

Sites of listed species are reported to Indiana DNR, Nature Preserves. Vouchers are currently held at the Indiana University Southeast Herbarium. Some collections are sent to the DNR, Nature Preserves for confirmation by Michael A. Homoya, DNR Botanist, as well as by George and Kay Yatskievych at the Missouri Botanical Garden. Methods and criteria for listing Indiana's extirpated, endangered, threatened, and rare vascular plant species have been discussed by Aldrich et al. (1986).

POSSIBLE STATE RECORDS AND NOTEWORTHY COLLECTIONS

Achyranthes japonica (Miq.) Nakai (Amaranthaceae). Japanese Chaff Flower. A native of Asia which was introduced into Kentucky

(Gleason & Cronquist 1991). Small, shrubby plants, common along the upper floodplain slopes and banks of the Ohio River in the Indiana counties across from Louisville, Kentucky. Colonies extend back from the river and down river to the west. We consider "W.E. Thomas 470" from Crawford County the state record voucher.

Egeria densa Planch. (Hydrocharitaceae). Brazilian Water-weed. The state record, "W.E. Thomas 294," was collected in shallow water from Wyandotte Lake in the Harrison-Crawford State forest. An aquarium plant "that has become naturalized in various localities in the United States, primarily in the southeast" (Beal & Thieret 1986).

Clematis terniflora DC. (= *C. dioscoreifolia* Levi. & Vaniot) (Ranunculaceae). A native of Japan which appears occasionally with other vines along the Ohio River.

Fatoua villosa (Thunb.) Nakai (Moraceae). Hairy Crabweed, Mulberry Weed. Reported in *Flora of North America* (3):390.1997, but neither the voucher nor the author of the Indiana report was identified (K. Yatskievych pers. commun.). The Floyd County record, "Maxwell *s.n.*, 9/20/1995" validates the FNA report and can be listed as the state record voucher. A native of tropical Asia, two plants were collected in a New Albany yard in 1995. We know of no other reports in Indiana.

Humulus japonicus Siebold & Zucc. (Moraceae). Japanese Hops. An introduced native of East Asia. The vine is fairly common along roadsides next to the Ohio River.

Lespedeza bicolor Turcz. (Fabaceae). Bicolor Lezpedeza. Two small shrubs were found in the open field northeast of the old black powder plant at Charlestown State Park. Reported for Indiana in the *Proceedings of the Indiana Academy of Science*, 1953. The state record voucher collected by "Buser & Ahles *s.n.*" in Fountain County is probably in the University of Illinois Herbarium at Urbana-Champaign.

Liparis loeselii (L.) Rich. (Orchidaceae). Loesel's Twayblade. Found by Homoya in Jackson County on the Muscatatuck National Wildlife Refuge (1990). Thomas discovered a population of about 14 plants distributed in the lower end of a seep in Charlestown State Park off Trail 1. This Clark County population represents a further southern extension as suggested by Homoya (1993).

Melothria pendula L. (Cucurbitaceae). Creeping Cucumber. This vine was considered state extirpated (SX) until three sites were discovered by Thomas in Charlestown State Park in 2000. The vines seemed to be thriving in spite of being in competitive habitats: a roadside ditch, an open disturbed area, and the edge of the escarpment over Fourteen Mile Creek. Since 2000 the Creeping Cucumber has been found in numerous sites throughout Charlestown State Park. It has also been found in roadside ditches in Floyd County along the lower edges of the knobs beside the Ohio River floodplain.

Mentha x rotundifolia (L.) Huds. (Lamiaceae). Apple Mint. A possible hybrid for southern Indiana. Several other collections have been made of this mint (Yatskievych 2000). Its occurrence in the state may be more common than the current reports indicate.

Orthodon dianthera (Buch.-Ham.) Hand. Mazz. (= *Mosla dianthera* (Buch.-Ham. ex Roxb.) Maxim) (Lamiaceae). A native of East Asia, established in moist places in the southeast (Gleason & Cronquist 1991). Recorded for Indiana in Kartesz & Meachan's 1999 Synthesis without a reference (K. Yatskievych pers. commun.). Homoya was aware of a site in the Harrison-Crawford State Forest and made a collection 26 September 1997 in Harrison County. The Homoya collection is the state record and "W.E. Thomas 36," a second collection. Thomas reports the population expanding in open areas left by logging at two different sites in Harrison-Crawford State Forest. This may be another species moving up from Kentucky and becoming common in southern Indiana.

Packera anonyma (A.W. Wood) W.A. Weber & Á. Löve (= *Senecio anonymous* A. Wood, *S. smallii* Britt.) (Asteraceae). Occasional on thin soil over a limestone ledge in Charlestown State Park. Perhaps a recent range expansion phenomenon (Homoya & Hedge 1990). Indiana Watch List (Yatskievych 2000).

Papaver dubium L. (Papaveraceae). Garden Poppy. The collection "Maxwell 3166." is the second report of the Garden Poppy along a different railroad track in Clark County. About 15 plants were noted along the tracks about 2½ km. north of highway I-265 along State Road 62 in 1998, but this second colony did not persist probably because of herbicide

Table 1.—State and county records. Abbreviations for species status are: SE = State Endangered, ST = State Threatened, SR = State Rare. Plant records without a number are indicated by *s.n.* = (*sine numero*) without a number, followed by the collection date.

Species	Family	County, collector and number
Acalypha gracilens A. Gray	Euphorbiaceae	Clark, Maxwell 3351
Acalypha deamii (Weath.) Ahles, (ST)	Euphorbiaceae	Harrison, W.E. Thomas 38
Achyranthes japonica (Miq.) Nakai	Amaranthaceae	Clark, W.E. Thomas 511; Crawford, W.E. Thomas 470; Floyd, W.E. Thomas 536
Aesculus pavia L.	Hippocastanceae	Floyd, Maxwell 3075
Albizia julibrissin Duraz.	Mimosaceae	Floyd, Boha & Chanley 8
Alisma subcordatum Raf.	Alismaceae	Harrison, R. Schoen *s.n.*, 21 July 1972
Amaranthus retroflexus L.	Amaranthaceae	Floyd, Weatherholt 1526
Ambrosia trifida L.	Asteraceae	Harrison, R. Schoen 59
Ammania robusta Heer & Regel	Lythraceae	Floyd, W.E. Thomas 28
Amorpha fruticosa L.	Fabaceae	Floyd, Schoen *s.n.*, 2 June 1972
Amsonia tabernaemontana Walter	Apocynaceae	Harrison, R. Schoen *s.n.*, 11 May 1972
Aquilegia canadensis L.	Ranunculaceae	Floyd, Weatherholt 880
Artemisia annua L.	Asteraceae	Clark, W.E. Thomas 30; Floyd, W.E. Thomas 635
Artemisia ludoviciana Nutt.	Asteraceae	Harrison, R. Schoen *s.n.*, 24 August 1972
Artemisia vulgaris L.	Asteraceae	Floyd, Maxwell 1280
Asclepias purpurascens L.	Asclepiadaceae	Floyd, S. Newman 113
Asplenium bradleyi D.C. Eaton (SE)	Aspleniaceae	Crawford, W.E. Thomas 159
Avena sativa L.	Poaceae	Floyd, Weatherholt 1561
Buglossoides arvense (L.) I.M. Johnst. (= *Lithospermum arvense* L.)	Boraginaceae	Floyd, Maxwell 3484
Capsella bursa-pastoris (L.) Medik.	Brassicaceae	Floyd, J.A. Kruer Jr. 90
Cardamine hirsuta L.	Brassicaceae	Floyd, Maxwell 2543
Cardamine angustata O.E. Schultz (= *Dentaria heterophylla* Nutt.)	Brassicaceae	Harrison, W.E. Thomas 157
Clematis terniflora D.C.	Ranunculaceae	Floyd, W.E. Thomas 17
Diodia virginiana L. (ST)	Rubiaceae	Floyd, W.E. Thomas 1; Clark, W.E. Thomas 335; Crawford, W.E. Thomas 435
Draba brachycarpa Nutt. ex Torr. & A. Gray	Brassicaceae	Clark, W.E. Thomas 156
Egeria densa Planch.	Hydrocharitaceae	Crawford, W.E. Thomas 294
Enemion biternatum Raf. (= *Isopyrum biternatum* (Raf.) Torr. & Gray)	Ranunculaceae	Floyd, Weatherholt 799
Equisetum arvense L.	Equisetaceae	Floyd, Weatherhold 845
Erianthus alopecuroides (L.) Ell.	Poaceae	Harrison, R. Schoen 47
Erythronium albidum Nutt.	Liliaceae	Floyd, Weatherholt 794
Erythronium americanum Ker Gawl.	Liliaceae	Floyd, Gohmann & Weatherholt 32
Fatoua villosa (Thunb.) Nakai	Moraceae	Floyd, Maxwell *s.n.*, 9/20/1995
Gymnocladus dioica (L.) K. Koch.	Caesalpiniaceae	Floyd, N.A. Myers 11
Holosteum umbellatum L.	Caryophyllaceae	Clark, Maxwell & Thomas 3197
Houstonia pusilla Schoepf	Rubiaceae	Clark, W.E. Thomas 62; Floyd, Maxwell 3483; Harrison, W.E. Thomas 58
Humulus japonicus Siebold & Zucc.	Moraceae	Clark, Reported by the Adams (1993); Crawford, W.E. Thomas 23; Floyd, W.E. Thomas 285
Hypochaeris radicata L.	Asteraceae	Clark, W.E. Thomas 340
Iva annua L. (= *I. ciliata* Willd. in Deam (1940))	Asteraceae	Clark, W.E. Thomas 18; Crawford, W.E. Thomas 125; Harrison, W.E. Thomas 495
Kickxia elatine (L.) Dumort	Scropulariaceae	Clark, Gilbert 105
Koelreuteria paniculata Laxm.	Sapindaceae	Floyd, F. Bierman 119

Table 1.—Continued.

Species	Family	County, collector and number
Lespedeza bicolor Turcz.	Fabaceae	Clark, Maxwell 3373
Linaria vulgaris Mill.	Scropulariaceae	Clark, K.D. Bledsoe 80
Lindernia dubia (L.) Pennell var. *anagallidea* (Michx.) Cooperr.	Scropulariaceae	Floyd, W.E. Thomas 291; Harrison, W.E. Thomas 295
Lindernia dubia (L.) Pennell var. *dubia*	Scropulariaceae	Crawford, W.E. Thomas 24; Harrison, W.E. Thomas 131
Linum usitatissimum L.	Linaceae	Floyd, Maxwell 3183
Liparis loeselii (L.) Rich.	Orchidaceae	Clark, Maxwell & Thomas 3498
Melothria pendula L.	Cucurbitaceae	Clark, Maxwell 3355; Floyd, Maxwell 3447
Mentha x rotundifoia (L.) Huds.	Lamiaceae	Crawford, W.E. Thomas 112; Floyd, W.E. Thomas 371
Muscari botryoides (L.) Mill.	Liliaceae	Harrison, R. Schoen *s.n.*, 28 April 1972
Nasturtium officinale R. Br.	Brassicaceae	Harrison, R. Schoen *s.n.*, 11 June 1972
Nothoscordum bivale (L.) Britton (SR)	Liliaceae	Crawford, W.E. Thomas 182
Oenothera biennis L. (= *O. pycnocarpa* Atkinson & Bartlett)	Onagraceae	Harrison, R. Schoen 36
Oenothera laciniata Hill	Onagraceae	Floyd, J.A.M.(? Illegible) 8
Oenothera pilosella Raf.	Onagraceae	Floyd, K. Heavrin 103
Osmorhiza longistylis (Torr.) D.C.	Apiaceae	Harrison, R. Schoen *s.n.*, 15 May 1972
Oxalis illinoensis Schwegman (SR)	Oxalidaceae	Clark, Maxwell 3432
Packera anonyma (A.W. Wood) W.A. Weber & Á. Löve	Asteraceae	Clark, Maxwell & Thomas 3492
Panicum nitidum Lam.	Poaceae	Floyd, R. Schoen 14
Pellaea glabella Mett. ex Kuhn	Adiantaceae	Crawford, W.E. Thomas 185
Penstemon digitalis Nutt. ex Sims	Scropulariaceae	Floyd, Hobson 96
Phleum pratense L.	Poaceae	Floyd, Barger & Weatherholt 72
Phlox divaricata L.	Polemoniaceae	Floyd, F. Gohmann 23
Platanthera lacera G. Don	Orchidaceae	Clark, Maxwell & Thomas observed
Platanus occidentalis L.	Platanaceae	Clark, Maxwell 1706
Polygonum arenastrum Jord. ex Boreau	Polygonaceae	Harrison, W.E. Thomas 42
Polygonum lapathifolium L.	Polygonaceae	Floyd, G. Doyle 7
Pyrrhopappus carolinianus (Walter) D.C.	Asteraceae	Clark, W.E. Thomas 5; Harrison, W.E. Thomas 272
Ranunculus repens L.	Ranunculaceae	Floyd, Weatherholt 1012
Ratibida columnifera (Nutt.) Woot. & Standl.	Asteraceae	Floyd, Maxwell 3337
Robinia hispida L.	Fabaceae	Clark, S. Newman 61
Rubus occidentalis L.	Rosaceae	Harrison, Conrad 71
Rumex obtusifolius L.	Polygonaceae	Clark, Megraw 40
Salix babylonica L.	Salicaceae	Floyd, Chambers 51
Salix exigua Nutt. (= *S. interior* Rowlee)	Salicaceae	Floyd, Bennett 2
Selaginella eclipes Buck	Selaginellaceae	Clark, Maxwell 3420; Floyd, Maxwell 3476
Sibara virginica (L.) Rollins	Brassicaceae	Clark, Maxwell 3478
Sida spinosa L.	Malvaceae	Harrison, S. Lawton 88
Solanum sarachoides Sendtner	Solanaceae	Clark, Maxwell 3367
Spermacoce glabra Michx.	Rubiaceae	Floyd, W.E. Thomas 3
Spiranthes vernalis Engelm. & A. Gray (SE)	Orchidaceae	Clark, W.E. Thomas 332
Thlaspi arvense L.	Brassicaceae	Harrison, W.E. Thomas 160
Tragia cordata Michx. (SR)	Euphorbiaceae	Clark, observed by Homoya
Typha angustifolia L.	Typhaceae	Floyd, Weatherholt 1573
Viola lanceolata L.	Violaceae	Harrison, R. Schoen *s.n.*, 19 May, 1972
Woodsia obtusa (Spreng.) Torrey	Aspleniaceae	Clark, Maxwell 1534

spraying along the right-of-way. The collection "W.E. Thomas 271," along State Road 31 north of Speed, indicates the initial Clark County colony is persisting and perhaps expanding in the much wider strip between State Road 31 and the railroad track. Its occurrence in the state may be more common than reports indicate.

Platanthera lacera (Michx.) G. Don. (Orchidaceae). Green Fringed Orchid. This orchid is the most common *Platanthera* in Indiana (Homoya 1993), but is primarily found in the northern part of the state. A single plant was discovered by Thomas in the edge of a wet depression off Jersey Avenue east of the current Charlestown State Park boundary in an area expected to become annexed to the park. A photo voucher was taken 6 July 2002. This report indicates a range extension to the south as predicted by Homoya (1993). Indiana Watch List (Yatskievych 2000).

Pycnanthemum muticum (Michx.) Pers. (Lamiaceae). Mountain Mint. The state record collector, K.F. Duffy, reports colonies in full sun, in poor clay soil next to a pond in a Harrison County hardwood forest. Deam (1940) excludes the species, citing reports, but no specimens. Schneck's report (Deam 1940) for the Lower Wabash Valley is probably correct. Mohlenbrock & Ladd (1978) show distribution in Wabash County, Illinois, next to the Wabash River.

Duffy sent a collection, "K.F. Duffy *s.n.*, 21 September 2002," to Kay Yatskievych at the Missouri Botanical Garden for verification; and this becomes the Indiana voucher specimen for the species and the county record. Our specimen, "K.F. Duffy 1," is a later collection from the same population.

Selaginella eclipes Buck (Sellaginellaceae). Small Spikemoss. First thought to be *S. apoda* (L.) Fern.(SE), later determined by Homoya as *S. eclipes*, and also confirmed by G. Yatskievych. Occasional to locally common in Charlestown State Park.

Solanum sarachoides Sendter (Solanaceae). Hairy Nightshade. A Gastony collection, "Gastony *s.n.*, 17 July 1985, Fee Lane and Highway 46 Bypass, Monroe County, Bloomington, was discovered in the Indiana University Herbarium. This may be the state record. The Gastony collection . . . "was apparently an escape to a roadside that was some distance from the experimental field" (K. Yatskievych pers. commun.). The experimental field refers to where Dr. Charles B. Heiser, Jr., of Indiana University carried out some of his well-known hybridization work with sunflowers, nightshades and domestic plants. Heiser (1969) refers to using *Solanum sarachoides* in his crosses to rediscover the "wonderberry."

The "Maxwell 3367, 25 August 2000," collection from Charlestown State Park would be a second report. This introduced weed was found in a disturbed area of the park and is persisting. However, it does not show the invasive capacity of other introduced species in the park.

INDIANA STATE ENDANGERED (SE)

Asplenium bradleyi D.C. Eaton (Aspleniaceae). Bradley's Spleenwort. Noted in Dubois County, 23 October 1984, by Homoya & Abrell (1986). "W.E. Thomas 159" was collected from a population of 13–15 plants scattered on several sandstone boulders in an open area near Wyandotte Cave in the Harrison-Crawford State Forest.

Gentiana villosa L. (Gentianaceae). Striped Gentian. Deam (1940) reports two collections from southeast Harrison County in black and white oak woods. A Harrison County site was located during a DNR conducted survey for the Indiana Natural Heritage Program. A fragment, "W.E. Thomas 37," was collected in the Harrison County part of the Harrison-Crawford State Forest in a cedar woods along Cold Friday Road. Thomas reports a scattered population of about 30–40 plants in the woods. These sites are in Harrison County, but we cannot be sure they are the same.

INDIANA STATE THREATENED (ST)

Acalypha deamii (Weath.) Ahles. (= *A. virginica* L. var. *deamii* Weath.) (Euphorbiaceae). Mercury. Michael Homoya identified this species in October 1994, in the Jenny Lind Run area of the Indiana Army Ammunition Plant. A few plants were also found on the south bank of Fourteen Mile Creek near the Ohio River. Over the last few years sizable populations were found by Thomas in Floyd County, along the Ohio River in New Albany, and Harrison and Crawford Counties. Maxwell ("Maxwell 3409, 10/19/2000") found a population of about 100 plants on a Fourteen Mile Creek sand bank near the north bridge

in what is now Charlestown State Park. Additional plants have turned up in the park along Trails 1, 2 and 3. It now appears so common in our area new sites are not reported to DNR, Nature Preserves.

Diodia virginiana L. (Rubiaceae). Buttonweed. Collected from large colonies along the Ohio River in Floyd and Crawford Counties, and also found along the wet perimeter of Deam Lake in Clark County. See Homoya et al. (1995) for additional locations. Now believed common along the Ohio River.

INDIANA STATE RARE (SR)

Gentiana flavida A. Gray (*G. alba* Muhl.) (Gentianaceae). Yellowish Gentian, Pale Gentian. An Indiana University Southeast Herbarium collection, "C.W. Henson 43, 19 Sept. 1975," from the Harrison-Crawford State Forest in a dry, oak woods clearing, was determined *Gentiana villosa*. Reexamination, as suggested by Homoya, showed it to be *Gentiana flavida*. The exact county distribution is unknown.

Northoscordum bivalve (L.) Britton (Liliaceae). Crow Poison. A colony of perhaps 60 plants was found on a low rock bluff along State Road 62 within Harrison-Crawford State Forest, west of Wyandotte Lake.

Oxalis illinoensis Schwegman (Oxalidaceae). Illinois Woodsorrel. Two large Clark County populations were pointed out by Homoya about 1994 on the wooded floor of Lick Creek Ravine, in what is now Charlestown State Park. At that time, one population contained about 100 individuals and the other 500–1000. Since then several additional clusters have been found in the park along with a population of about 2000 individuals spreading down from the upper bluff on the east side of Lick Creek towards Fourteen Mile Creek. Two colonies, one with perhaps 100 plants, the other smaller with perhaps 20 plants, were found by Thomas in Crawford County. Both these sites were within the Harrison-Crawford State Forest along the upper tributaries of Dry Run Creek.

The Illinois Woodsorrel has leaflets shallowly lobed with green margins; the Big Yellow Woodsorrel (*Oxalis grandis* Small) has leaflets deeply lobed with purplish-brown margins (Yatskievych 2000). Other characteristics are similar except the Illinois Woodsorrel has a small tuberous root (Homoya pers. commun.). We believe the Illinois Woodsorrel is more common than the Big Yellow Woodsorrel in our south central Indiana area.

Spiranthes vernalis Engelm. & A. Gray (Orchidaceae). Spring Ladies' Tresses, Grass-leaved Ladies' tresses. Observed by Thomas & Howell Curtis, Charlestown State Park in a disturbed area north of Trail 1 parking. "W.E. Thomas 332" serves as a Clark County voucher.

Tragia cordata Michx. (Euphorbiaceae). Heart-leaved Noseburn. Homoya identified this vine in the rocky woods, north of the west little bluestem glade in Charlestown State Park.

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DISTRIBUTION OF FRESHWATER SPONGES AND BRYOZOANS IN NORTHWEST INDIANA

David K. Barnes: Department of Biology, Purdue University – North Central; Westville, Indiana 46391-9528 USA

Thomas E. Lauer: Aquatic Biology and Fisheries Center, Department of Biology, Ball State University, Muncie, Indiana 47306-0440 USA

ABSTRACT. Current biogeographical distributions of freshwater sponges (Porifera) and bryozoans (Ectoprocta and Entoprocta) are poorly known in Indiana. Although seemingly ubiquitous in many aquatic communities, neither group has received much notoriety in this state, nor the midwestern United States. In an initial effort to begin the systematic taxonomic distribution of these groups in Indiana, this study identified 2 sponges and 13 bryozoans in northwest Indiana lakes. The recent appearance of the zebra mussel (*Dreissena polymorpha*) in the Great Lakes has focused new attention on the sessile benthic communities, as significant changes in benthic community structure are expected with the introduction of this exotic species. Without knowing the historical and current distribution of native invertebrates, such as the sponges and bryozoans, it is not possible to identify changes in community composition over time.

Keywords: Porifera, Ectoprocta, Entoprocta, freshwater bryozoan, freshwater sponge

The distribution and taxonomy of freshwater sponges (Porifera: Spongillidae) in North America has not received widespread attention, although some notable works exist for the entire United States (Penney & Racek 1968), and regions surrounding the Great Lakes (Potts 1887; Smith 1921; Old 1931; Jewell 1935; Neidhoefer 1940; Eshleman 1950; Ricciardi & Reiswig 1993). Ricciardi & Reiswig (1993) found 15 different species and suggested that additional taxa could be identified in eastern Canada with further study. Sponges may be more ubiquitous than the available scientific literature indicates; and the absence or limited ranges of some species may not reflect their true zoogeographical distribution, but rather a lack of observation (Frost 1991). We have only been able to find six published records of sponge distributions in Indiana (Evermann & Clark 1920; Kintner 1938; Early et al. 1996; Lauer & Spacie 1996; Early & Glonek 1999; Lauer et al. 2001), and these are limited in scope.

Although bryozoans (Ectoprocta and Entoprocta) are widely distributed in epibenthic and littoral communities (Rogick 1934, 1957; Bushnell et al. 1987; Ricciardi & Lewis 1991), little is known about their zoogeographical status. There are 25 species known in North American freshwater (Wood 2001; Smith 1992), and the distribution of many of these species is thought to be ubiquitous (Bushnell 1974). In the Great Lakes region, some taxonomic records exist (Brown 1933; Rogick 1934; Bushnell 1965a, 1965b, 1965c; Maciorowski 1974; Ricciardi & Reiswig 1994). However, no published documentation could be found for this taxonomic group in Indiana except for these recent studies in the northern part of the state (Wood 1996; Lauer et al. 1999; Last & Whitman 1999/2000).

Without knowing the historical and current distribution of native sponges and bryozoans, it is not possible to identify changes in community composition over time. Rapid modification in the quantity and diversity of benthic species has been observed in the Great Lakes with the appearance of non-indigenous species, such as the zebra mussel (*Dreissena polymorpha*). These invasions have threatened the biotic integrity of native organisms and negatively affected the ecosystem stability of sessile benthic communities (Hebert et al. 1991; Mills et al. 1994).

The objectives of this study were to determine the biogeographical distribution of fresh-

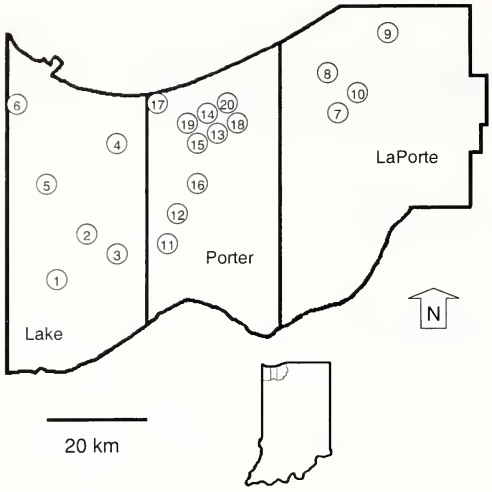

Figure 1.—Lakes studied in Lake, Porter and LaPorte Counties, Indiana. Numbers indicate lake location and correspond to the lake names: 1. Cedar; 2. Francher; 3. Holiday; 4. Lakeland; 5. Lemon; 6. Wolf; 7. Clear; 8. Pine; 10. Saugany; 11. Big Bass; 12. Eliza; 13. Flint; 14. Long; 15. Loomis; 16. Louise; 17. Minnehaha; 18. Round; 19. Spectacle; 20. Wahob.

water sponges and bryozoans in the lakes of Lake, Porter and LaPorte counties, Indiana (excluding Lake Michigan), and to identify the water quality habitats of both bryozoans and freshwater sponges where they are found.

METHODS

Twenty lakes in three northwest Indiana counties were sampled for freshwater sponges and bryozoans during the summer of 1998 (Fig. 1). The lakes sampled were public, freshwater natural lakes remnant from the geological activity that occurred during the Pleistocene glaciation approximately 12,000 years ago (reviewed by Hutchinson 1957). The size and maximum depths of the lakes varied (Table 1). In each lake, a single collection effort occurred in June–July and typically included 2–4 man-hours of shallow water wading and snorkeling at each site. For small lakes, the collection effort encompassed the entire littoral area, while for larger lakes, the collection sites included all available habitat types. No sampling was performed at the lake outlets. Scuba diving was used in deeper waters when warranted. Sticks, plants, rocks, and other similar hard substrate were examined for the sponges and bryozoans. Collections entailed picking or scraping the organisms from the substrate, or removing the organism with the substrate still attached. Sponge identification is based largely on spicule morphology and care was taken to collect specimens bearing gemmules, as well as somatic tissue. Similarly, bryozoan identification typically requires the inspection of statoblasts in addition to the vegetative colony, and both were collected when possible. Most species are large enough to observe with the naked eye, but a hand lens was used to validate colonies that were small or non-descript. Samples were preserved immediately upon collection with 70% ethyl alcohol, and returned to the laboratory for taxonomic identification.

Laboratory preparation of sponge spicules (megascleres, microscleres, and gemmoscleres) was performed using an acid digestion, followed by washings in water and ethyl alcohol. The alcohol was evaporated, and the spicules were mounted in Permount. This technique creates a permanent mount and allows the microscopic examination of the spicules as described in Pennak (1989). Species were identified using the taxonomic keys of Pennak (1989), Frost (1991), and Ricciardi & Reiswig (1993).

Bryozoan colonies and statoblasts were examined using a binocular microscope at 10× magnification and identified using the taxonomic keys of Pennak (1989), Wood (1989, 2001), Ricciardi & Reiswig (1994) and Wood (1996).

Water quality measurements were taken at each location at the time of biological sampling to characterize the habitat where bryozoans and sponges were found. Physical and chemical parameters measured were: temperature, pH, Secchi, conductivity, hardness, calcium, magnesium, total dissolved solids, and dissolved oxygen. Chemistry analysis was completed within 6 h of collection using Hach Company methodology (Hach Chemical Company 1988, 1990) for oxygen (DO), hardness, calcium, magnesium, and total dissolved solids (TDS). A digital Marine Systems meter was used for pH measurements, while conductivity and temperature were taken on site using a Hach Model 44600 conductivity meter.

RESULTS

Thirteen bryozoans (3 classes, 7 families) were identified in the 20 study lakes (Table 2)

Table 1.—Lake basin morphometry, and physical and chemical measurements of the water found at each of the 20 sampling locations during 1998. Water samples were collected at the time of sponge and bryozoan sampling.

		Max.	Max.	Water quality parameter								
County/Lake	Date	size (ha)	depth (m)	Temp °C	pH S.U.	Secchi m	D.O. mg/l	Hard. mg/l	Ca mg/l	Mg mg/l	Cond. µmhos/cm	TDS mg/l
Lake												
1. Cedar	7/10	316	5.0		8.7	4.0	7.8	74	29.6	10.8	911	456
2. Francher	7/10	4.0	12	27.3	8.6	1.0	8.2	78	31.2	11.4	386	193
3. Holiday	7/6	77	5.5	31.0	7.8	0.3	6.9	76	30.4	11.1	394	198
4. Lakeland	7/8	2.8	10	27.3	8.7	4.0	7.8	74	29.6	10.8	911	456
5. Lemon	7/10	13	4.1	27.3	7.3	0.2	6.5	29	11.6	4.2	416	212
6. Wolf	7/13	156	2.5	31.6	8.8	1.8	8.0	65	26.0	9.5	289	147
LaPorte												
7. Clear	6/22	43	3.5	28.9	9.4	1.0	9.0	60	24.0	8.8	653	331
8. Pine	6/24	228	16	29.8	9.0	2.0	9.3	39	15.6	5.7	293	148
9. Saugany	7/3	30	20	28.4	8.5	5.0	7.5	47	18.8	7.0	432	218
10. Stone	6/23	51	11	28.4	8.7	3.0	8.0	54	21.6	7.9	259	136
Porter												
11. Big Bass	7/6	28	3.4	28.1	7.8	0.3	4.0	71	28.4	10.4	338	170
12. Eliza	7/15	18	14	28.0	9.4	0.4	10.7	61	24.4	8.9	267	134
13. Flint	6/29	36	20	29.4	8.7	1.5	7.0	67	26.8	9.8	351	175
14. Long	6/30	26	8.2	26.9	7.8	1.0	5.8	27	10.8	4.0	221	111
15. Loomis	6/30	25	17	29.1	8.4	0.5	5.8	53	21.2	7.8	361	181
16. Louise	7/16	92	7.6	28.2	8.4	1.6	7.5	75	30.0	11.0	334	168
17. Minnehaha	7/13	8.9	11	29.6	8.6	5.0	8.3	78	31.2	11.4	550	276
18. Round	6/29	0.7	5.5	29.1	7.5	2.1	6.0	41	16.4	6.0	352	219
19. Spectacle	6/30	12	3.0	29.5	8.5	1.4	5.8	57	22.8	8.4	412	201
20. Wahob	6/29	8.0	15	28.8	7.5	3.0	6.1	44	17.6	6.4	271	136

Table 2.—Sponges and bryozoans found in Lake, Porter, and LaPorte counties, Indiana. The lake site number corresponds to the corresponding lake number in Figure 1.

	Lake number
Bryozoans	
Phylum Ectoprocta	
Class Phylactolaemata	
Family Fredericellidae	
Fredericella indica Annandale 1909	2, 3, 6, 17
Fredericella browni (Rogick 1945)	8, 10
Family Plumatellidae	
Hyalinella punctata (Hancock 1850)	2, 17
Plumatella casmiana (Oka 1907)	3
Plumatella emarginata Allman 1844	3
Plumatella fungosa (Pallas 1768)	1
Plumatella orbisperma (Kellicott 1882)	9
Plumatella nitens Wood 1996	1, 6, 12
Plumatella reticulata Wood 1988	1, 2, 3, 8, 10, 12, 13, 16
Family Pectinatellidae	
Pectinatella magnifica (Leidy 1851)	3, 11, 12, 18, 20
Family Cristatellidae	
Cristatella mucedo Cuvier, 1798	7, 8, 9, 10, 13, 16, 18, 20
Class Gymnolaemata	
Family Paludicellidae	
Paludicella articulata (Ehrenberg 1831)	13, 17
Phylum Entoprocta	
Family Urnatellidae	
Urnatella gracilis Leidy 1851	1
Sponges	
Phylum Porifera	
Family Spongillidae	
Eunapius fragilis Leidy 1851	3, 13, 20
Spongilla nr. *aspinosa* Potts 1880	16, 18, 20

and represent the first published account of these species from Indiana. Twelve of them are in the Phylum Ectoprocta and follow the classification scheme outlined by Wood (2001), while a single species, *Urnatella gracilis*, is in the Phylum Entoprocta.

Two freshwater sponge species, *Eunapius fragilis* and *Spongilla aspinosa*, were also found. *Eunapius fragilis* has been found previously in the Indiana waters of Lake Michigan (Lauer & Spacie 1996), while *Spongilla aspinosa* is a new record for Indiana.

Physical and chemical measurements (Table 1) provided some reference to the environmental limits of the bryozoans and sponges. Although the samples for measurement were collected at the 1 m depth, they provided some indication of the range of tolerance of these species. This is particularly true of some species, such as *Cristatella mucedo*, that were found at multiple sites.

DISCUSSION

Finding 12 ectoproct bryozoan species (2 classes, 6 families) in a relatively limited three-county area in northwest Indiana was unexpected. As a comparison, Bushnell (1965a) sampled 122 sites in 48 counties in Michigan over four years and found 13 species, while Wood (1989) found 13 species from 60 locations in Ohio, and Ricciardi & Reiswig (1994) found 14 species from 80 localities in eastern Canada. Other investigators found similar numbers of species (Lake Erie, Rogick (1934); Ottawa River, Ricciardi & Lewis (1991); Louisiana, Everitt (1975)), although some recent changes in ectoproct classifications (see Wood 1996, 2001) may alter the number of species. Our findings suggest that the 20 lakes surveyed in Lake, Porter and LaPorte Counties have a diverse environmental character and support a wide variety of ec-

toproct bryozoan species despite the small biogeographical area surveyed.

The most common ectoproct species by distribution were *Fredericella indica*, *Plumatella reticulata*, *Pectinatella magnifica*, and *Cristatella mucedo* (Table 2). The first three species are widely distributed in Ohio (Wood 1989) and Michigan (Bushnell 1965a), while *C. mucedo* is found throughout North America, but less frequently (Bushnell 1965a; Smith 1985; Wood 1989; Ricciardi & Reiswig 1994). *Pectinatella magnifica* colonies can grow to a large size (> 50 cm) and are readily found as a gelatinous matrix, particularly late in the summer. Although our specimens were not this large (10–30 cm), they were readily seen during collections. Wood (1989) states that this species also has a wide tolerance for water quality characteristics, but appears to be thermophilic, as growth of statoblasts into colonies is limited below 20° C. Water temperatures at the time of our collections in all 20 lakes were above this limit and did not appear to be inhibiting growth of this species.

Fredericella indica, *Plumatella casmiana*, *Plumatella emarginata*, *Plumatella repens* and *Paludicella articulata* are widely distributed in the midwestern United States and North America (Brown 1933; Bushnell 1965a; Wood 1989; Ricciardi & Lewis 1991; Ricciardi & Reiswig 1994); and their occurrences in northwest Indiana were not unexpected.

Despite the apparent widespread distribution throughout the U.S. (Wood 2001), *Urnatella gracilis* was found only in Cedar Lake in our study. Wood (2001) suggested this species does well in areas where there are extensive water movements, either flowing water or the shallow area of large lakes. Cedar Lake was the largest lake in the study (316 ha) and is shallow (maximum depth 5 m) (Indiana Department of Natural Resources 1966).

Two species of sponges, *Eunapius fragilis* and *Spongilla* nr. *aspinosa*, were found in 5 of the 20 lakes (Table 2). *Eunapius fragilis* is a common species in the midwestern U.S. (Old 1931; Jewell 1935; Lauer & Spacie 1996). It has been found on all continents and climates and is described as truly cosmopolitan by Harrison (1974). As might be expected, this species exhibits a wide tolerance to environmental conditions (summarized by Harrison 1974), well within the water quality measurements we observed (Table 1). The sponge has a preference for growing in areas where the canal system is not clogged by siltation (Potts 1887), with growth typically being enhanced on the underside of hard substrates (Lauer & Spacie 1996). *Spongilla* nr. *aspinosa* is far less common than *E. fragilis* and has been reported only from waters that have low pH (Potts 1887; Eshleman 1950; Ricciardi & Reiswig 1993). Identification of *S.* nr. *aspinosa* was tentative, as gemmules are required for positive confirmation; and we could not find any. Gemmules are over-wintering structures and typically used as a protection against adverse conditions. However, *S.* nr. *aspinosa* is environmentally tolerant and rarely produces gemmules as part of its life history strategy (Potts 1887). Although some question in identification remains for this species, this is probably the correct name for this organism.

Associating water quality parameters with specific organism species was a bit difficult from our results with the limited sampling effort of this survey. However, these lakes are generally characterized as being eutrophic, with extensive anthropogenic inputs.

Both the sponges and the bryozoans in this study represent new records for Indiana. These records should not be considered range extensions or changes in habitats where they were found, but rather, simply an initial assessment of the sponge and bryozoan biogeographical distributions in Indiana. Many of these species are found throughout the geographical region, and might be expected. Considering the relative ease with which these organisms can be found, the paucity of studies on these organisms in Indiana or even nationwide does not suggest they have a limited distribution, but rather that they have not been studied. Our limited mid-summer sampling effort may have also missed some species that are present due to seasonal fluctuations or variations in the growth and development of these organisms. More intense and complete works on the subject, e.g., Wood (1989) and Ricciardi & Reiswig (1993, 1994), support this hypothesis. This study identified the biogeographical distribution of these little known groups in northwest Indiana, and by doing so, initialized an effort to more fully understand the significance and ecological importance of sponges and bryozoans to this region.

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PSEUDOTREMIA REYNOLDSAE, A NEW SPECIES OF TROGLOBITIC MILLIPED (DIPLOPODA: CHORDEUMATIDA: CLEIDOGONIDAE), WITH A SYNOPSIS OF THE CAVERNICOLOUS MILLIPEDS OF THE HOOSIER NATIONAL FOREST IN INDIANA

Julian J. Lewis: J. Lewis & Associates, Biological Consulting, 217 West Carter Avenue, Clarksville, Indiana 47129 USA

ABSTRACT. A bioinventory of caves of the Hoosier National Forest in south-central Indiana has resulted in the collection of 12 milliped taxa, including a new species, *Pseudotremia reynoldsae*, which is described and illustrated. This species is known from only one isolated cave in the Patoka River drainage in Crawford County. *Pseudotremia reynoldsae* is structurally most similar to *P. burnsorum*, a troglobite occurring in caves of the Mosquito Creek drainage in Harrison County. Three other subterranean chordeumatidan species inhabit the Hoosier National Forest: *Pseudotremia salisae*, *Pseudotremia indianae* and *Conotyla bollmani*. Other native species found were *Euryurus leachii leachii*, *Scytonotus granulatus*, *Pseudopolydesmus* sp., *Cambala minor*, *Abacion tesselatum* and *Petaserpes* sp. Two exotic milliped species occur in the Hoosier National Forest, *Oxidus gracilis* and *Ophyiulus pilosus*.

Keywords: Cave millipeds, *Pseudotremia*, Indiana, Hoosier National Forest, Patoka River

Until 1997 only two species of cavernicolous millipeds of the genus *Pseudotremia* were known from Indiana, *Pseudotremia indianae* Hoffman (1958) from the Blue River area and *P. nefanda* Shear (1972) from Clark County. Biological sampling in over 400 Indiana caves (e.g., Lewis 1998; Lewis et al. 2002) has demonstrated a diverse subterranean milliped fauna. Hoffman and Lewis (1997) described *P. conservata* from Seven Springs Cave, Harrison County, and Lewis (2000) added five more new species of *Pseudotremia* from caves in Harrison and Crawford Counties: *P. blacki*, *P. burnsorum*, *P. cookorum*, *P. purselli* and P. salisae.

In 2000 a systematic bioinventory of the caves of the Hoosier National Forest (HNF) was begun. The HNF extends from the Ohio River north to an area east of Bloomington, Indiana. Much of the forest lies within the physiographic unit known as the Crawford Upland (Powell 1961), where cavernous limestone strata are capped with sandstone. There are presently 136 caves known to occur on HNF lands and a like number of privately-owned caves are known to exist within the proclamation boundary of the national forest.

The first species of *Pseudotremia* discovered in the HNF was found in two caves at the Hemlock Cliffs Special Area on the Little Blue River drainage (Lewis 1998) and subsequently described as *P. salisae* (Lewis 2000). Two more species of *Pseudotremia* have now been discovered within the boundary of the HNF, *P. indianae* and an undescribed species that represents the ninth member of the genus to be described from southern Indiana.

In my previous work on *Pseudotremia* (Lewis 2000) I followed the terminology concerning gonopod structures established by Shear (1972). In the light of the present understanding of gonopod anatomy, Shear (pers. commun. 2002) has suggested a new interpretation followed herein. In *Pseudotremia* the gonopods develop only from the eighth pair of legs, with the ninth pair reduced in size and playing little role in sperm transfer. Of the structures evolving from the eighth pair of legs in chordeumatidans, the telopodite is either entirely lost (as in *Pseudotremia*), or if present, is not a functional part of the gonopod (Shear 2000). The structures called the telopodite by Shear (1972) and Lewis (2000) are

actually permanently extruded, sclerotized coxal glands fused at the bases, that are now termed syncolpocoxites; and the process arising from the fused base is termed the syncolpocoxal process (Shear pers. commun. 2002). The structures referred to by Shear (1972) as colpocoxites are angiocoxites developed from the margin of the coxal gland pore and are divided into medial and lateral parts (Shear pers. commun. 2002).

METHODS

Between 2000–2002 a total of 104 caves was sampled within the Hoosier National Forest. The primary means of sampling the milliped fauna was pitfall traps using four ounce (approximately 160 ml) jars filled with 70% isopropyl alcohol and baited with Kraft™ limburger process cheese spread. To a lesser extent specimens were collected by hand or obtained through Berlese extraction of organic litter. Appendages were placed on temporary glycerin slide mounts and drawn using a Leica compound microscope with an optical drawing tube.

In the interest of cave conservation specific localities are not provided. Those in need of specific localities can apply for the information from the Indiana Cave Survey (www.caves.org/conservancy/ICS).

The spelling of the word milliped (rather than millipede) as explained by Hoffman (2000) is the usage preferred by North American specialists in the group dating back to its apparent origin with Cook in 1911 and is followed herein.

MILLIPEDS OF THE HOOSIER NATIONAL FOREST

Twelve taxa discovered to date in caves of the HNF are listed with the sampling sites recorded by county of occurrence. Many of the caves in which collecting has been conducted occur on national forest wilderness or special areas that have been designated as such for the protection of the ecosystems. In the following list the taxa are arranged by county, with caves sampled within designated forest service areas listed in the appropriate area.

ORDER CHORDEUMATIDA

Family Cleidogonidae

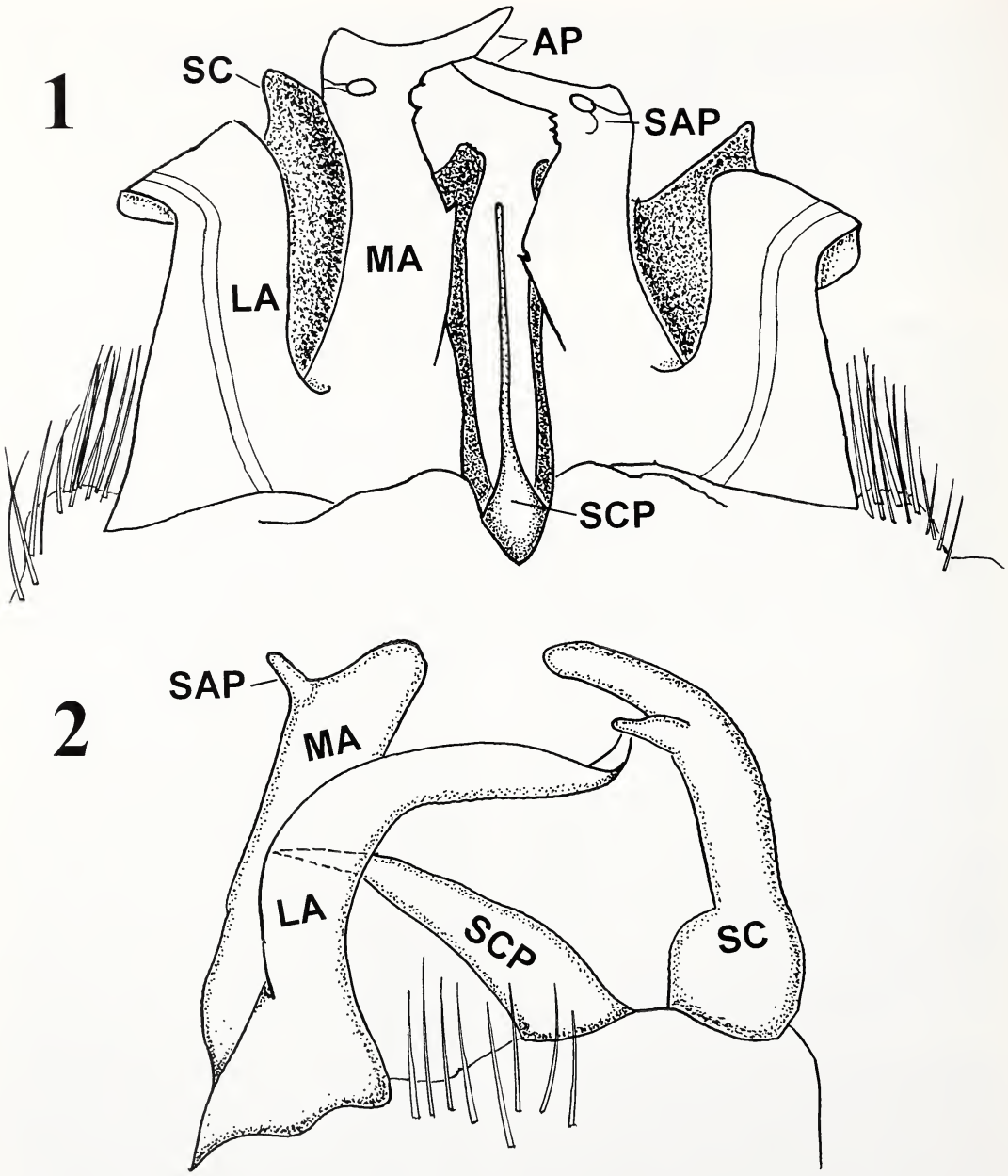
Pseudotremia reynoldsae new species

(Figs. 1–5)

Material examined.—*Crawford County:* Megenity Peccary Cave, south room, holotype ♂, 3 juveniles, 28 July 2001 (J. Lewis, Salisa T. Rafail); same locality, paratype ♂, 5 juveniles, 8 September 2001 (J. Lewis, Ron Burns). All material has been deposited in the Virginia Museum of Natural History, Martinsville, Virginia.

Diagnosis.—Moderately pigmented and eyed; anterior gonopods with syncolpocoxite mitten-shaped with prominent distolateral knob, process simple, entire, of saber type of Shear (1972); median angiocoxite with irregularly dentate medial margin, apical knob and process and reduced subapical process, lateral angiocoxite broad, entire. This species is most similar to *P. conservata* and *P. burnsorum* that occur in caves in southern Harrison County, and *P. nefanda* in caves in Clark County. *Pseudotremia reynoldsae* is immediately separable from this assemblage by the short subapical process on the median angiocoxite. *Pseudotremia reynoldsae* can furthermore be separated from all known *Pseudotremia* in Indiana by the presence of the dentate mesial margin of the median angiocoxite.

Description.—*Male:* Longest approximately 26 mm in length (coiled), maximum width 2.2 mm (7th segment); body overall medium brown with purplish suffusion, each metatergum with a prominent subovate reticulated spot bilaterally and scattered tubercles, adults notably more pigmented than subadults and juveniles, which appear more uniformly light beige. Head and antenna darker purple, sternum and legs light beige with purplish tinge. Eyes with 18–19 unpigmented ocelli within subtriangular, pigmented ocellaria. Antennae about 4.2 mm long, slender, 3rd segment longest, about 1.2 mm. Segmental paranota small to moderate in size, largest anteriorly, becoming indistinguishable from lateral striae in posterior segments. Lateral striae 11–13. Gonopods with syncolpocoxites separated by U-shaped cleft; process simple, tapering to a point, of the saber type of Shear (1972). Median angiocoxite, mesial margin irregularly dentate; apical round knob, two processes, the larger a blade-shaped apical structure crisscrossing in the cleft between the angiocoxites, the other a smaller subapical structure originating from the lateral side and extending anteriomesiad, not extending beyond mesial margin of angiocoxite. Lateral angiocoxite separated from median by a relatively deep, U-shaped cleft, broad, entire, simply curving.



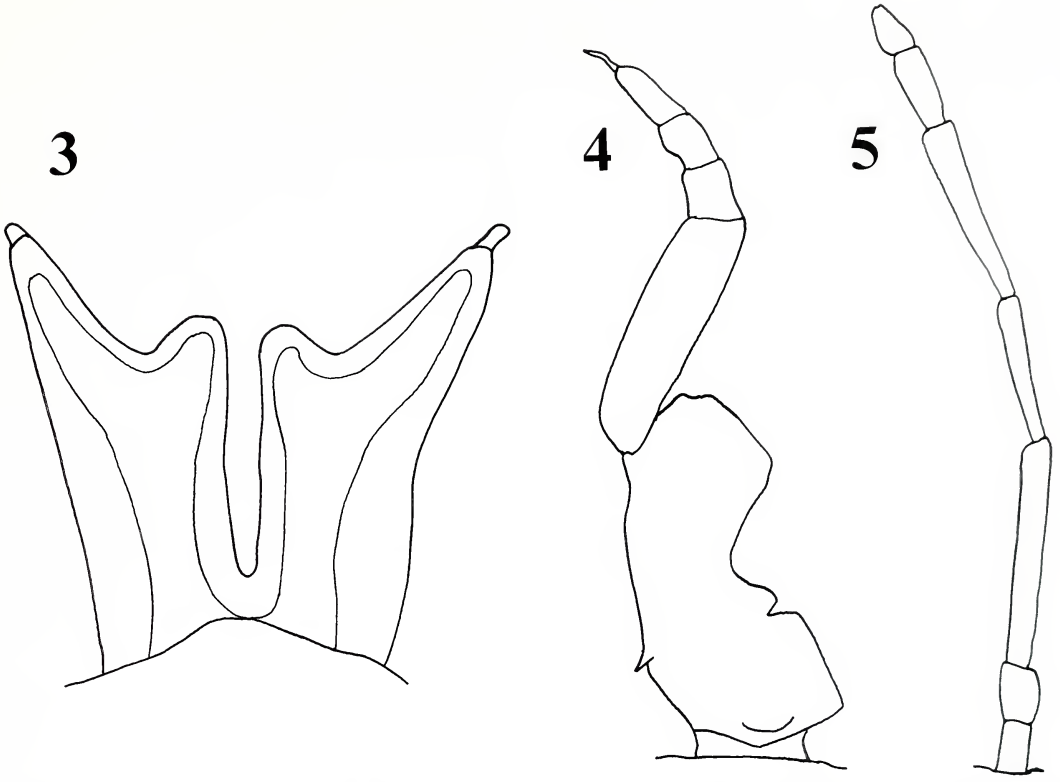
Figures 1–2.—***Pseudotremia reynoldsae*** new species, Megenity Peccary Cave, Crawford County, Indiana, holotype male. 1. Anterior gonopods, anterior aspect; 2. Anterior gonopods, lateral aspect. *Abbreviations:* SC = syncolpocoxite; SCP = syncolpocoxite process; LA = lateral branch of angiocoxite; MA = median branch of angiocoxite; AP = apical process; SAP = subapical process.

Leg 9 with basal spinous process on mesial margin, broadly rounded apophysis absent; distal 3 segments well formed and apparently fully functional, claw present.

Female: Adult unknown.

Etymology.—This species is named in honor of Kelle Reynolds, Forest Wildlife Biologist and Karst Coordinator for the Hoosier National Forest, U.S. Forest Service. The suggested vernacular name is the Reynold's cave milliped.

Habitat and range.—*Pseudotremia rey-*



Figures 3–5.—***Pseudotremia reynoldsae*** new species, Megenity Peccary Cave, Crawford County, Indiana, holotype male. 3. Syncolpocoxite, anterior aspect; 4. Leg 9; 5. Antenna.

noldsae is known only from Megenity Peccary Cave, which lies in the Patoka River drainage in south-central Indiana. This cave consists of 126 m of mapped passages formed in limestone and enlarged by collapse of the overlying sandstone. The cave is maze-like, comprised of a series of joint controlled passages that were apparently originally streamless; but it has been excavated for paleontological material (Richards 1994), and one area now contains flowing water. *Pseudotremia reynoldsae* specimens were taken on and around cheese-baited pitfall traps in the parts of the cave farthest from the entrance.

Pseudotremia salisae Lewis

HNF localities.—*Crawford County:* Enlow's Back Door Cave, Perfect Circle Cave, Salt Shake Rock Cave. Hemlock Cliffs Special Area: Sentinel Rock Cave, Pavey Cave.

Habitat and range.—Lewis (2000) listed the species from Heron and Mesmore Spring Caves in the HNF Hemlock Cliffs Special Area, in Crawford County, and Pipe Cave in Harrison County. This species is notably guanophilic and was found associated with raccoon droppings at every collection site.

Pseudotremia indianae Hoffman

HNF localities.—*Orange County:* Duggin's Spring Cave, Little Africa Pleasure Palace Cave, Springs Spring Cave, Wells Cave.

Habitat and range.—This milliped is the most highly cave-adapted of any of the Indiana species of *Pseudotremia* and is primarily an inhabitant of riparian habitat where it occurs on mudbanks adjacent to cave streams. It has previously been recorded from numerous caves along the Blue River drainage in Crawford, Harrison, Washington and Orange Counties (Shear 1972; Hoffman & Lewis 1997) and is known to have crossed the drainage divide eastward into the Buck Creek drainage (Lewis 2000). Duggin's Spring Cave lies in an area of the Blue River drainage that extends into the eastern edge of the HNF. The Springs Spring Cave collection is the first record of *P. indianae* extending past the western drainage

divide of Blue River, crossing into the Patoka River drainage. At Little Africa Pleasure Palace, *P. indianae* occurs in the headwaters of the East Fork of White River drainage on the same ridge as Bond Cave (inhabited by *Conotyla bollmani*).

Family Conotylidae
Conotyla bollmani (McNeill)

HNF localities.—*Lawrence County:* King/Bug Ear Cave, Carpenter Cave; Tincher Karst Special Area: Brick Pit, Burton Hollow Cave, Crystal Falls Cave, Fuzzy Hole, Gory Hole, Henshaw Bend Cave, Horse Bone Hole, HNF Swallowhole, JJ's Cave, Smith's Folly Cave, Tincher Hollow Cave, TRAC Cave, William's Cave. *Martin County:* Bluff House Cave, Redberry Cave; Gypsy Bill Allen Special Area: Gypsy Bill Allen Cave. *Monroe County:* Charles C. Deam Wilderness: Dead Possum Pit, Frog Pond Pot, Frog Pond Ridge Pit, Patton Cave. *Orange County:* Apple Cave, Beaver Attack Cave, Bond Cave, Dillon Cave, Eckleberger Cave; Paoli Experimental Forest: Garlow Spring Cave; Wesley Chapel Gulf Special Area: Elrod Cave, Wesley Chapel Gulf Cave; Springs Valley Recreation Area: Campground Cave, Not Our Area Cave, Tucker Dam Quarry Cave, Tucker Lake Spring Cave.

Habitat and range.—This milliped occurs in riparian habitats as well as streamless cave habitats like pit floors, particularly on rotting wood. It occurs in caves in the East Fork of White River drainage, from south-central Orange County through Lawrence into Monroe and Owen Counties (Hoffman & Lewis 1997). A record of a juvenile *Conotyla* from a cave on the Crosley State Fish & Wildlife Area in Jennings County is suggestive of this species, but remains unconfirmed. Shear (1972) cited a surface collection of this species, but provided no specific records, perhaps an allusion to the "near Wyandotte Cave" collection of *Scotherpes* (sic) *wyandotte* (= *Conotyla bollmani*). I have sampled over 190 caves in that area (Hoffman & Lewis 1997; Lewis 2000), and *C. bollmani* was never found, so it seems unlikely that this species occurs in the Blue River drainage.

ORDER SPIROSTREPTIDA
Family Cambalidae
Cambala minor Bollman

HNF localities.—*Crawford County:* Heron Cave, Megenity Peccary Cave. *Lawrence County:* Tincher Karst Special Area: Smith's Folly Cave.

Habitat and range.—This species is troglophilic and frequently found on raccoon droppings. In Indiana it has been recorded from caves in Clark, Crawford, Harrison, Lawrence, Orange and Washington Counties (Shelley 1979; Lewis 1996, 1998; Lewis et al. 2002), as well as surface records from Monroe, Posey and Putnam Counties (Bollman 1888). The range includes the south-central United States, from Oklahoma east into western Virginia.

ORDER POLYDESMIDA
Family Euryuridae
Euryurus leachii leachii (Gray)

HNF localities.—*Crawford County:* Megenity Peccary Cave; Hemlock Cliffs Special Area: Arrowhead Arch. *Lawrence County:* Tincher Karst Special Area: Smith's Folly Cave. *Martin County:* Dave's Dig Cave.

Habitat and range.—This species is a trogloxene and has been reported in Indiana from caves in Crawford, Harrison, Jennings, Lawrence, Martin and Orange Counties (Hoffman 1978; Lewis, 1995, 1998; Lewis et al. 2002). Bollman (1888) reported this species from surface habitats (as the synonym *E. erythropygus* per Hoffman 1999) in the additional counties of Benton, Clark, Hamilton, Howard, Putnam, Tippecanoe, Vigo and Washington. This species is found in the central United States in Illinois, Indiana, Kentucky and Ohio (Hoffman 1978, 1999).

Family Paradoxosomatidae
Oxidus gracilis (Koch)

HNF localities.—*Crawford County:* Carnes Mill Special Area: Carnes Mill Spring Cave. *Lawrence County:* Carpenter Cave; Tincher Karst Special Area: Henshaw Bend Cave, South Gardner Mine.

Habitat and range.—This milliped is an exotic of uncertain origin, although indications are that it probably came from Japan. It now occurs in a nearly world wide distribution in temperate climates and at higher elevations in the tropics. In North America it occurs in "astronomical numbers" in many parts of the southeastern U.S. (Hoffman 1999). In Indiana it has been reported from caves in Clark, Crawford, Harrison, Jennings, Lawrence, Orange and Washington Counties (Lewis

1995, 1996, 1998, 2002). Sampling of rotting logs in the Deam Wilderness Area, Monroe County, revealed that *Oxidus* outnumbered the native *Scytonotus* by a ratio of perhaps 50:1. *Oxidus* was not noted as present in Indiana at the time of Bollman (1888).

Family Polydesmidae

Scytonotus granulatus Say

HNF localities.—*Monroe County:* Charles C. Deam Wilderness: Patton Cave. *Orange County:* Paoli Experimental Forest: Garlow Spring Cave; Wesley Chapel Gulf Special Area: Wesley Chapel Gulf Cave.

Habitat and range.—This species is a trogloxene that usually occurs in leaf litter in caves. In Indiana it has been reported from caves in Crawford, Harrison, Jennings, Monroe, Orange and Washington Counties (Lewis 1995, 1998). The range of *S. granulatus* extends through the southeastern U.S. (Shelley 1993). Bollman (1888) characterized the species as abundant.

Pseudopolydesmus sp.

HNF localities.—*Martin County:* Dave's Dig Cave. *Orange County: Paoli Experimental Forest: Q1B3 Cave.*

Habitat and range.—Hoffman (1999) reported 12 species in this genus occurring across eastern North America. The localities cited represent collections of single juveniles taken in the entrance zone of the caves in deep leaf litter.

ORDER POLYZONIIDA

Family Polyzoniidae

Petaserpes sp.

HNF localities.—*Monroe County:* Charles C. Deam Wilderness: unnamed pit on Frog Pond Ridge. *Orange County:* Grease Gravy Special Area: Grease Gravy Cave; Springs Valley Recreation Area: Not Our Area Cave.

Habitat and range.—Shelley (1997) reported three species of *Petaserpes* from Indiana, including *P. bikermani* from Donaldson Cave and *P.* sp. from Sullivan Cave, both in Lawrence County. The HNF collections are comprised of juveniles that can not be identified to species.

ORDER CALLIPODIDA

Family Caspiopetalidae

Abacion tesselatum Rafinesque

HNF localities.—*Crawford County:* Enlow's Back Door Cave. *Orange County:* Grease Gravy Special Area: under rocks at spring associated with Grease Gravy Cave.

Habitat and range.—This species occurs from the Gulf Coastal Plain through the east-central U.S., where it is a common inhabitant of both native and urban environments (Shelley 1984).

ORDER JULIDA

Family Julidae

Ophyiulus pilosus (Newport)

HNF localities.—*Lawrence County:* King/Bug Ear Cave; Tincher Karst Special Area: Brick Pit, Fuzzy Hole, Garden Hose Pit, HNF Swallowhole, Pail Cave, Tincher Swallowhole Cave, Tincher Hollow Cave, Williams Cave. *Orange County:* Allen Cave; Paoli Experimental Forest: Garlow Spring Cave.

Habitat and range.—This species is an exotic probably native to Italy that is now widespread in the northeastern U.S. (Hoffman 1999). It frequents caves and is ubiquitous in leaf litter and detritus in caves of the Tincher Karst Special Area. It is also known from caves in Clark and Harrison Counties (Lewis 1996, 1998).

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AMPHIBIANS AND REPTILES FROM TWENTY-THREE COUNTIES OF INDIANA

Robert Brodman: Biology Department, Saint Joseph's College, Rensselaer, Indiana 47978 USA

ABSTRACT. Amphibians and reptiles from 23 counties, 13 in northern Indiana and 10 in southwestern Indiana, were surveyed from 1998–2001 using standardized call-survey, terrestrial and aquatic search-and-seize, and trapping methods. A total of 3514 populations of amphibians representing 33 of the 39 species known from Indiana was found at 1539 sites. The most common species, western chorus frogs (*Pseudacris triseriata*, spring peepers (*P. crucifer*), American toads (*Bufo americanus*), and green frogs (*Rana clamitans*), comprised 74% of the frog populations encountered, while smallmouth salamanders (*Ambystoma texanum*), tiger salamanders (*A. tigrinum*), unisexual salamanders of the *A. laterale*-complex, and red-backed salamanders (*Plethodon cinereus*) made up 67% of the salamander populations. Five species of amphibians with distributions within the areas that I surveyed, plains leopard frog (*R. blairi*), hellbender (*Cryptobranchus alleganiensis*), red salamander (*Pseudotriton ruber*), green salamander (*Aneides aeneus*), and northern dusky salamander (*Desmognathus fuscus*), were not found. I maximized observations of amphibian species richness by using combinations of techniques rather than a single method. All amphibian species were more abundant in wildlife preserves than along randomly chosen routes. Reptiles were encountered much less frequently than were amphibians; however, most of the methods were targeted towards detecting amphibian communities. I observed 286 reptiles representing 27 species at 129 sites.

Keywords: Amphibians, reptiles, survey, Indiana

Amphibian declines throughout the world (Barinaga 1990; Houlahan et al. 2000; Lannoo et al. 1994; Wake 1991) have emphasized the need for an inventory of species to determine how widespread declines have been and to serve as baseline data for long-term monitoring and future surveys to assess changes in the status of the herpetofauna. The US Central Division of the Declining Amphibian Population Task Force recommended atlas projects in each state.

The checklist is the fundamental record of an atlas project. From these checklists, atlas maps can be produced; however, for data to be compared among locations or time, effort must also be recorded. Measures of species seen per unit effort can be calculated and used to investigate regional relative abundance patterns, changes over time, and changes among species (Cyr & Larivee 1993). The lack of this information limits the usefulness of historical data on amphibians and reptiles in Indiana (Grant 1936; Minton 2001; Mittleman 1947; Simon et al. 2002; Swanson 1939).

The Indiana Declining Amphibian Population Task Force and the Indiana Department of Natural Resource Technical Advisory Committee on amphibians and reptiles initiated an Indiana Herpetofauna Atlas with the goal to conduct surveys throughout the state over a four-year period of time (1998–2001) to produce data on the presence and abundance of amphibians and reptiles with quantified effort. Here I report on my surveys of 23 counties in Indiana. My objectives are to: 1) determine the status of the herpetofauna and species status state-wide during a four year period; 2) determine past trends of the herpetofauna at certain sites with rich herpetological records; 3) provide baseline data on species richness, relative abundance and species status of amphibians and reptiles that can be used to determine population trends and begin long-term monitoring of certain populations; and, 4) analyze amphibian presence and abundance patterns among randomly chosen sites and non-randomly chosen preserves.

METHODS

Preserve sites were chosen non-randomly in representative regions and habitats of the state targeting those properties with historical records, high species diversity, rare or endangered species and prospects for long-term protection of

Figure 1.—Map of survey locations by county. Counties surveyed for amphibians and reptiles are labeled and shaded with diagonal lines. Small dots indicate counties with additional information on the abundance of amphibians from surveys conducted in the 1990s in the recent literature and from Indiana Department of Natural Resources.

habitat. I also surveyed random samples of sites in blocks representing each major region of the state's herpetofauna (Brodman 1998; Minton 2001; Smith & Minton 1957). Effort was equalized among visits to a given site and among blocks, but not among counties. From 1998–2001 I surveyed 56 preserve sites in 23 counties. Random samples of survey sites were selected from a random set of 50 national wetland inventory topographic maps 1:24,000 scale (NWI) from northern Indiana and 15 NWI maps from southwestern Indiana. I targeted wetlands that were within 200 m of secondary roads. Most were in agricultural fields and residential properties. I surveyed sites in Jasper, Benton, White, Pulaski, Newton, Huntington and Wabash Counties in 1998; Wabash, Miami, Huntington, Adams and Jay Counties in 1999; Sullivan, Greene, Owen, Monroe, Martin, Daviess, Dubois, Spencer, Floyd and Posey Counties

in 2000; and Newton, Pulaski, Starke, Fulton and Wells Counties in 2001 (Fig. 1). Of the 39 species of amphibian known from Indiana, my survey sites were within the range of every species except the ravine salamander-complex (*Plethodon richmondi/electromorphus*) (Highton 1999; Minton 2001). To simplify the situation of unisexual populations of salamanders with various hybrid chromosome combinations from *Ambystoma laterale*, *A. jeffersonianum*, or *A. texanum*, I will consider these as a single taxon and refer to them as unisexual salamanders.

I considered amphibian breeding-ponds within 0.5 km of each other to be a single population site. All sites were surveyed using standard methods of amphibian population monitoring (Heyer et al. 1994; Karns 1986). Estimates of population abundance were quantified by a combination of anuran call indices, density estimates and per effort catch. I conducted anuran call surveys on at least three occasions (late March–mid April, late April–late May, early June–early July) at wetlands throughout the study areas targeted each year. Relative abundance was estimated on an ordinal scale while effort was measured as the number of km surveyed. I used time constrained search-and-seize using seines and dip nets for aquatic species, terrestrial/cover object searches, minnow traps for aquatic species, and visual sweeps of appropriate habitat for basking animals, eggs and larvae. Effort was measured as the number of person-hours spent using each method. Minnow traps were used to determine relative abundance by recording the number of animals caught per trap-day.

The primary methods employed in this survey were intensive directed sampling and opportunistic sampling. Directed sampling is sampling in areas that clearly should contain amphibians. Opportunistic sampling includes night-time road cruising with periodic (every 0.5 km) auditory sampling, visual sweeps through candidate areas searching for terrestrial animals, and overturning logs near wetland habitats. Small wetlands were dip-netted and seined thoroughly and systematically; but for larger wetlands, shallow edges were randomly sampled at several locations. Egg-mass densities were determined by quadrat sampling (see Brodman 1995). Terrestrial salamanders were monitored by numbers caught or seen per person-hours of hand search-and-seize methods. Numbers of egg masses, larva and adult animals caught or seen per person-hours of dip netting, seine sampling and opportunistic sampling give an indication of density and survey effort.

Table 1.—Effort by method in each county surveyed. Data are km of call route (CR), terrestrial survey person-hours (TS), aquatic survey person-hours (AS), the number of minnow trap-days (MT) and total effort (TE) for each county.

County	CR	TS	AS	MT	TE
Southern Indiana					
Floyd	45	15	14	0	74
Greene	50	13	14	15	92
Sullivan	71	18	18	15	122
Daviess	55	15	15	15	100
Owen	39	55	55	0	149
Monroe	42	60	65	5	172
Spencer	42	16	13	0	71
Martin	39	14	12	0	65
Posey	56	21	25	30	132
Dubois	97	14	15	5	131
Northern Indiana					
Wells	39	12	13	12	76
Adams	82	13	13	0	108
Jay	103	15	16	0	134
Huntington	122	14	15	0	151
Wabash	166	22	40	0	228
Miami	108	20	20	0	148
Benton	333	18	18	0	369
White	50	12	12	0	74
Pulaski	69	42	42	38	191
Newton	150	43	48	20	261
Starke	24	35	35	14	108
Jasper	830	60	60	108	1058
Fulton	21	18	18	34	91
Total	2631	565	596	311	4103

I used auditory frog call surveys for the assessment of amphibian breeding intensity. Relative population abundance was categorized using the Karns (1986) ordinal index of breeding chorus intensity on a scale of 0–5. Salamander and reptile relative abundance were also categorized on an ordinal scale from 0–5 using the following formulas:

Aquatic survey
= Ln (5 × captures/person-hour)

Minnow traps
= Ln (35 × captures/trap-day)

Terrestrial survey

Table 2.—Summary of amphibian abundance. Data are overall abundance (AB), number of sites present (SP), the percentage of counties (%C) or preserves (%P) present, and the ratio of new county records to absences from historic counties (CR). Data are presented from random sites, non-randomly chosen preserves and totals for the state.

	Random		Preserves			Total			
Species	AB	SP	AB	SP	%P	AB	SP	%C	CR
Frogs									
Chorus	1916	650	577	219	86	2493	869	100	3:0
Spring peeper	1656	506	574	200	68	2230	706	87	7:0
Eastern gray	663	246	205	76	32	868	322	48	3:1
American toad	616	252	199	87	34	814	339	83	6:3
Cricket	242	92	236	85	30	478	177	74	3:7
Green	221	123	201	97	64	422	220	100	11:0
Cope's gray	208	96	182	86	34	390	182	52	5:0
Fowler's toad	241	102	132	69	39	373	171	74	4:3
Bullfrog	114	64	125	73	48	239	137	100	11:0
Northern leopard	106	64	94	45	36	200	109	57	2:2
Southern leopard	12	8	120	42	29	132	50	43	0:1
Wood	13	8	42	15	20	55	23	35	5:2
Crawfish	0	0	3	3	2	3	3	13	1:5
Spadefoot	2	1	1	1	2	3	2	9	1:1
Pickerel	0	0	2	1	2	2	1	4	0:3
Salamanders									
Smallmouth	15	6	110	39	39	125	45	74	8:3
Tiger	38	15	41	18	23	79	33	43	7:5
Unisexual	9	5	52	19	28	61	24	48	8:0
Red-backed	0	0	37	17	16	37	17	43	0:5
Blue-spotted	0	0	27	18	7	27	18	13	2:1
Spotted	0	0	23	13	13	23	13	22	1:4
Two-lined	1	1	16	6	11	17	7	26	1:5
Marbled	0	0	13	7	9	13	7	26	1:3
Jefferson	2	1	11	4	5	13	5	22	1:0
Lesser siren	0	0	11	8	7	11	8	17	0:2
Zigzag	0	0	10	6	9	10	6	17	0:2
Slimy	0	0	10	4	5	10	4	13	1:6
Eastern newt	0	0	8	6	5	8	6	13	2:2
Streamside	0	0	8	2	4	8	2	4	0:0
Cave	0	0	4	1	2	4	1	4	0:4
Four-toed	0	0	3	2	2	3	2	9	1:1
Longtail	0	0	3	2	2	3	2	4	0:6
Mudpuppy	0	0	2	2	4	2	2	4	0:2
Total	6075	2240	3082	1273	—	9157	3514	—	96:86

$$= \text{Ln}\ (450 \times \text{captures/person-hour})$$

All calculated values below 1 were rounded up to 1 if at least one animal was encountered and all values greater than 5 were rounded down to 5. A total effort of 2631 km of call survey routes, 565 person-hours of terrestrial surveys, 596 person-hours of aquatic surveys and 311 minnow trap-days was conducted in this study (Table 1).

RESULTS

Amphibians.—I detected a total of 3514 populations at 1539 sites in the 23 counties that I surveyed. Data for each species are summarized in Table 2. There was a significant and strongly positive correlation between the relative abundance of species and the number of sites that each species was present (Pearson correlation, $r = 0.994$; $p < 0.001$).

I found 15 of the 16 species of frogs known from Indiana. The only species absent was the plains leopard frog (*Rana blairi*). I found 18 of Indiana's 23 salamander species. I did not encounter the hellbender (*Cryptobranchus alleganiensis*), red salamander (*Pseudotriton ruber*), green salamander (*Aneides aeneus*), northern dusky salamander (*Desmognathus fuscus*) and the ravine salamander-complex. I found 96 new county records, and there were 86 instances of a species absent from a county with a historical record (Table 2). The species with the most new county records were the bullfrog (*Rana catesbeiana*), green frog (*R. clamitans*), spring peeper (*Pseudacris crucifer*), smallmouth salamander (*Ambystoma texanum*), tiger salamander (*A. tigrinum*), and unisexual salamander. The species with the most absences from counties with historical records were the crawfish frog (*R. areolata*), cricket frog (*Acris crepitans*), longtail salamander (*Eurycea longicauda*), and northern slimy salamander (*Plethodon glutinosus*).

The most abundant species of frogs were the western chorus frog (*P. triseriata*), spring peeper, eastern gray treefrog (*Hyla versicolor*) and American toad (*Bufo americanus*). When combined, these four species accounted for two-thirds of all frog populations encountered. The western chorus frog, green frog and bullfrog were the only species found in all 23 counties. The most abundant salamanders were the smallmouth salamander, tiger salamander, red-backed salamander (*Plethodon cinereus*), and unisexual salamander. These species represent two-thirds of the total salamander abundance that I detected.

Amphibians were relatively more abundant and had greater species richness at the non-randomly chosen preserves compared to sites from random blocks (Table 2). All 33 species of amphibians encountered in this survey were found at the preserves, but two species of frog, the pickerel frog (*R. palustris*) and crawfish frog, and most of the salamander species (78%) were not found at the random sites. After weighing the abundance and number of sites by effort, amphibians were found to be about three times more common and abundant at the preserves than at random sites. If I exclude the four most abundant frog species, then the other species were about seven times as likely to be found or heard in preserves than along a random route. The greatest differences for species encountered at least 20 times were for the southern leopard frog (*R. sphenocephala*) and wood frog (*R. sylvatica*).

The number of sites that each species was found in each county is summarized in Table 3. Floyd and Owen Counties had the most amphibian species of any county, while Benton County had the fewest. The counties with the greatest number of new county records were Huntington, Miami, Pulaski and Sullivan, while Martin, Dubois, Greene, and Monroe Counties had the most species with past records absent (Table 3). Counties with the greatest number of sites with amphibians present were Fulton, Posey, Daviess and Floyd, whereas Adams, Dubois, Huntington, and Benton had the fewest (Table 3).

I compared the field methods to determine if any were more efficient or effective. During aquatic surveys seines yielded a mean of 3.7 species per site and 45.0 animals per person-hour, dip-nets yielded a mean of 4.9 species per site and 17.4 animals per person-hour, and minnow traps yielded a mean of 3.0 species per site and 2.4 animals per trap-day. Terrestrial and aquatic search-and-seize and visual sweeps yielded a mean of 4 species per site and 0.2 animals per person-hour. Although seining collected the greatest number of amphibians per person-hour and aquatic dip-nets produced the greatest species richness per person-hour, the combination of all four techniques was necessary to maximize species richness. For example, I encountered the lesser siren (*Siren intermedia*) and mudpuppy (*Necturus maculosus*) only in minnow traps, whereas the eastern spadefoot toad (*Scaphiopus holbrooki*) and crawfish frog were encountered only during night-time call surveys.

Reptiles.—I found reptiles at 129 sites in 19 of the 23 counties (Table 4). Just three species, painted turtle (*Chrysemys picta*), common garter snake (*Thamnophis sirtalis*) and eastern box turtle (*Terrapene carolina*)) made up more than half (53%) of the 286 reptiles found in this survey and accounted for 9 of the 13 new county records. The six-lined racerunner (*Cnemidophorus sexlineatus*), bullsnake (*Pituophis melanoleucus*), and eastern hognose snake (*Heterodon platirhinus*) were also relatively common; however, I observed each in only two counties in northwest Indiana. I found turtles in 65% of the counties, snakes in 61% of the counties and lizards in

Table 3.—Summary of the presence and abundance of amphibians in each county surveyed. Data are the number of sites for each species in each county, new county records (+) and absences (−) from counties with prior records. Counties are coded as follows: Floyd = fd, Posey = po, Dubois = du, Martin = ma, Spencer = sp, Monroe = mo, Owen = ow, Daviess = da, Sullivan = su, Greene = gr, Adams = ad, Jay = jy, Wells = we, Huntington = hu, Wabash = wa, Miami = mi, Fulton = fu, Benton = be, White = wh, Starke = st, Pulaski = pu, Newton = ne, and Jasper = ja.

	Counties								
Species	fl	po	du	ma	sp	mo	ow	da	su
Frogs									
Spring peeper	30	0	20+	8	5+	16	18	15	48
Chorus	26	43	38	25	4	8	14	28	34
Northern leopard	0−	0	0	0	0	0−	0	0	0
Pickerel	1	0	0	0−	0	0−	0	0	0
Southern leopard	5	18	4	4	5	2	1+	2	7
Crawfish	0	0	0−	0−	1	1	0	0−	1+
Eastern gray	0	0	0	0	0	0	0	0	0
Cope's gray	31	30+	2	21	9	3	11	20+	9
American toad	5	3	0−	14	0−	7	3	1	0
Fowler's toad	5	12	5+	3	10	1	6	14	7
Spadefoot	1	0	0	0−	0	0	1+	0	0
Cricket	19	20	3	2	15	3	0−	34	31
Green	8	9	2	4	4+	4	5	14+	6+
Bullfrog	8	10	1	2	3	3+	1	24+	7+
Wood	1+	1+	0	0−	0	4	3	1+	0
Salamanders									
Tiger	0−	0	0	0	0	0−	0−	2	1+
Smallmouth	1	16	1	1	1+	0−	1	4	3+
Streamside	2	0	0	0	0	0	0	0	0
Jefferson	0	0	0	1	0	1	1	0	1
Unisexual	1+	1+	0	0	0	1	2+	0	0
Blue-spotted	0	0	0	0	0	0	0	0	0
Spotted	2	7	0	0−	1	0−	0−	0	0
Marbled	1	2	0−	0−	0−	1	1+	1	1
Eastern newt	1+	0	0	0	0	1	0−	0	0
Lesser siren	0	3	0−	0	0	0	0	0−	1+
Mudpuppy	0	2	0	0	0	0	0	0	0
Red-backed	1	1	0−	0−	0−	3	4	0−	1
Zigzag	1	1	0	0−	0	1	3	0	0
Slimy	0−	1	0−	0−	0−	0−	1	0	0
Four-toed	0	0	0	0	0	1	0	0	0
Two-lined	2	0	0−	1	0−	1	1	0	0
Longtail	0−	0	0−	0	0−	2	0−	0	0
Cave	0−	0	0	0−	0−	0−	1	0	0

30% of the counties surveyed. I observed about 10% of the reptiles while driving. Fifteen of the specimens encountered were road-killed and another 13 were live animals seen along roadsides.

Reptiles were encountered much less frequently than were amphibians, and all but 10 of the reptiles observed were in preserves. Because several of the methods targeted amphibians, and most of the terrestrial searches were in preserves, no direct comparisons can be made between amphibians and reptiles or reptiles in preserves and those in random sites.

DISCUSSION

Amphibians.—The strong correlation between the abundance and number of sites present among species confirms that the trend first observed in Jasper County (Brodman & Killmurry 1998) is a state-wide phenomenon.

Table 3.—Extended.

Counties													
gr	ad	jy	we	hu	wa	mi	fu	be	wh	st	pu	ne	ja
14+	2+	0−	0	4+	65	35+	48	17+	36	10	38	66	211
8	12	41	6+	13+	67	33	23	88	35	13	36	63	171+
0	2	8	1+	4	15	12+	18	3	2	6	8	12	18
0	0	0	0	0	0	0−	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0	0	0
0−	0	0	0	0	0	0	0	0−	0	0	0	0	0
0	0−	7	0	1+	21	43	31	28+	27	5+	30	29	100
10	0−	0−	0	0−	0−	0−	0−	0−	0−	0−	10	5+	21
0−	1	4	3	4+	19	22+	7	77	16+	2+	21+	48	82+
0−	0	0−	0	1+	2	11+	8	0	6	1	12	16	53
0	0	0	0	0	0	0	0	0	0	0	0	0	0
23+	1+	8	2	0−	2	2+	0−	0−	4	0−	7	0−	1
4+	2+	4	2+	6+	15	8+	24	10+	9+	3	26+	6	46
4+	4	6	2	3+	3+	5+	17+	2+	2	2	7+	6+	19
0	0	0	0	0	0−	0	3+	0	0	3	7+	0	0
2+	3	1+	1+	1+	2	0−	1	0	1	0−	1+	2+	21
2+	3	2	1	1+	4	0−	0	0	1+	1+	3+	0	0−
0	0	0	0	0	0	0	0	0	0	0	0	0	0
0	0	0	1+	0	0	0	0	0	0	0	0	0	0
0	2+	3+	1+	2+	5+	0	0	0	0	3	3+	0	0
0	0	1+	0	0	0	0	0−	0	0	0	8+	0	9
0	0	0	0	0	1	1	0	0	0	0	1+	0	0
0	0	0	0	0	0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0−	0	0	0	0	0	4+
0−	0	0	0	0	0	0	0	0	0	0−	1+	0	3
0	0	0	0	0	0	0	0−	0	0−	0	0	0	0
1	0	0	0−	0	2	2	0	0	0	2	0	0	0
0−	0	0	0	0	0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0	0	2+	0	0	0−
0	0	0	0	0	0−	0	0	0	0	2+	0	0	0
0−	0	0	0	0−	1	1+	0	0	0−	0	0	0	0
0−	0	0	0	0	0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0	0	0	0	0	0

This suggests that presence-absence data are sufficient in determining relative abundance of amphibians on a county or state regional-level. Statewide monitoring programs need not use abundance indices that are more prone to human error and observer bias (Mossman et al. 1998). In studies using large numbers of people to conduct call surveys, observers agreed on presence or absence of species much more often than when they agreed on index values (Bishop et al. 1997; Hemesath 1998; Kline 1998; Shirose et al. 1997). This study also supports the finding that combining call surveys with time-constrained search-and-seize methods increases the accuracy of determining the relative abundance of frogs and toads (Kline 1998).

My finding that amphibians are disproportionately more abundant in preserves than along random routes brings into question

Table 4.—Summary of the presence and abundance of reptiles in each county surveyed. Data are the total number of animals seen or captured for each species in each county, and number of sites present (SP) for each species. An asterisk indicates a new county record. Counties are coded as follows: Floyd = fd, Posey = po, Dubois = du, Martin = ma, Spencer = sp, Monroe = mo, Owen = ow, Daviess = da, Sullivan = su, Greene = gr, Adams = ad, Jay = jy, Wells = we, Huntington = hu, Wabash = wa, Miami = mi, Fulton = fu, Benton = be, White = wh, Starke = st, Pulaski = pu, Newton = ne, and Jasper = ja.

	Counties																				
Species	fl	po	du	ma	sp	mo	ow	da	su	gr	ad	jy	wa	fu	be	wh	st	pu	ne	ja	SP
Turtles																					
Snapping	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	2	3	4	8
Painted	0	12	2	5	1	0	0	1+	1+	1+	0	0	3	14	0	2	2	0	15	38	26
Blanding's	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	4	2
Map	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	1
Red-eared slider	0	5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	4
Eastern box	8	3	3+	0	1+	0	0	2+	1+	0	0	0	0	0	0	0	0	3+	0	1	4
Ornate box	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	4
Spiny softshell	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	4	4
Lizards																					
Slender glass	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	2
Six-lined racerunner	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	9	2	8
Five-lined skink	0	1+	1	0	1+	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	6
Broad-headed skink	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	1
Snakes																					
Racer	1	1	1	0	0	0	0	1	0	0	0	0	0	0	0	0	2	0	2	1	7
Northern ringneck	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	1+	0	0	0	2
Black rat	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
Western fox	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0	2	0	4
Eastern hognose	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	11	2	8
Prairie king	0	0	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	1
Northern copperbelly	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
Northern water	3	1	2	0	0	1	1	0	1+	0	0	0	1	0	0	0	0	0	1	2	11
Rough green	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
Bull	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	12	4	12
Brown	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	1	4	4
Plains garter	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0	0	0	1	4
Eastern ribbon	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	4	3
Common garter	0	0	3+	1	0	0	1	1	0	0	1	1	3	0	0	4	0	0	8	6	18
Cottonmouth	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1

Table 5.—Amphibian relative abundance from recent surveys in 49 counties of Indiana. Data include the number of sites with populations present (SP) and percentage of counties present (%C) from my study, recent DNR reports and published papers.

Species	SP	%C
Frogs	4241	
Chorus	1004	92
Spring peeper	829	84
American toad	447	88
Eastern gray	418	43
Green	357	96
Bullfrog	225	94
Cricket	209	59
Fowler's toad	205	65
Cope's gray	204	49
Northern leopard	170	53
Wood	75	39
Southern leopard	63	39
Pickerel	28	22
Spadefoot	4	8
Crawfish	3	6
Plains leopard	0	0
Salamanders	623	
Smallmouth	114	53
Tiger	72	39
Red-backed	64	51
Blue-spotted	63	20
Cave	54	9
Unisexual	38	29
Spotted	34	47
Two-lined	29	29
Eastern newt	25	31
Zigzag	22	22
Northern slimy	19	27
Longtail	17	20
Hellbender	15	6
Marbled	14	24
Jefferson	13	24
Streamside	8	12
Lesser siren	8	8
Four-toed	6	6
Mudpuppy	4	6
Northern dusky	2	2
Ravine-complex	1	1
Green	1	1
Red	0	0

whether random routes give an accurate depiction of amphibian distribution and abundance. Coupled with the negative effects of roadway traffic (Ashley & Robinson 1996; Fahrig et al. 1995) and road salt on amphibian mortality (Turtle 2000), surveying wetlands that are only within 200 m of roads may be biased towards underestimating amphibian abundance and overestimating declines.

Regardless of these concerns, I have provided a baseline data set on amphibian relative abundance that can be repeated. Future studies should involve long-term monitoring of some of the preserves and random routes surveyed herein.

Reptiles.—With the exception of painted turtles observed basking in numerous habitats, reptiles were infrequently encountered during this survey. Although 286 reptiles were observed in 23 counties, I do not feel that this sample size is robust enough for the same analysis that was done on the amphibian data. This is primarily because my methods were targeted for amphibians and reptiles that use wetlands. However, after spending over 1000 person-hours in the field and finding so few reptiles, the apparent rarity of reptiles may be real.

Historical trends.—Population trends observed by comparing my results to those from previous surveys can be made within limitations (Lannoo et al. 1994). Although past methods were not standardized or effort quantified, ranked relative abundance of species can be compared. Several of my survey areas had been surveyed in past decades. Minton (1998) surveyed sites in Jasper, Pulaski, Benton and Floyd Counties from 1948–1993. Grant (1936) reported amphibians and reptiles observed at Jasper-Pulaski Fish and Wildlife Area and Starke County from 1931–1934. Swanson (1939) collected amphibians and reptiles from Jasper, Pulaski, Martin, Monroe, Posey, Greene, Daveiss, and Dubois Counties. Mittleman (1947) reported notes on amphibians and reptiles collected from 1945–1946 in Monroe, Owen, and Martin Counties.

Grant (1936) reported the racer (*Coluber constrictor*), eastern ribbon snake (*Thamnophis sauritus*), common garter snake, six-lined racerunner and eastern hognose snake as common in Jasper-Pulaski Fish and Wildlife Area; and he reported the painted turtle, spotted turtle (*Clemmys guttata*), ornate box turtle (*Terrapene ornata*) and Blanding's turtle (*Emydoidea blandingii*) as common in Jasper-Pulaski Fish and Wildlife Area and in Starke County. Swanson (1939) considered the slender glass lizard (*Ophisauris ventralis*), six-lined racerunner, Blanding's turtle, spotted tur-

tle, ornate box turtle, and fox snake (*Elaphe vulpina*) to be common at Jasper-Pulaski Fish and Wildlife Area. I do not consider any of these species to be common in these areas today.

Swanson (1939) listed the smallmouth salamander, red-backed salamander, longtail salamander, Fowler's toad (*Bufo fowleri*), chorus frog, crawfish frog, eastern fence lizard (*Sceloporus undulatus*), five-lined skink (*Eumeces fasciatus*), worm snake (*Carphophis amoena*), northern ringneck snake (*Diadophis punctatus*), racer, eastern hognose snake, common kingsnake (*Lampropeltis getulus*), northern water snake (*Nerodia sipedon*), copperbelly water snake (*Nerodia erythrogaster*), red-bellied snake (*Storeria occipitomaculata*), rough green snake (*Opheodrys aestivus*), copperhead (*Agkistrodon mokasen*) and timber rattlesnake (*Crotalus horridus*) to be common in southern Indiana. Among these species I would only consider the chorus frog, Fowler's toad, and smallmouth salamander as currently common species in southwestern Indiana.

Mittleman (1947) considered the zigzag salamander (*Plethodon dorsalis*), two-lined salamander (*Eurycea cirrigera*), cricket frog, gray treefrog (*Hyla versicolor* and *H. chrysoscelis*), bullfrog, green frog, and queen snake (*Regina septemvittata*) to be "exceedingly common" in Indiana. Today only green frogs, the two gray treefrog species, and cricket frogs are common, and the later is common only in the southern part of its range.

Minton (1998) reported several species that declined during his lifetime. The cave salamander (*Eurycea lucifuga*), longtail salamanders, and zigzag salamander were plentiful in Floyd County but declined in the 1970s. The plains leopard frog, crawfish frog, and Blanding's turtle were common in Benton County in the early 1950s but did not occur in 1993. The cricket frog and blue-spotted salamander were common at Jasper-Pulaski Fish and Wildlife Area in the 1940s and 1950s but declined in the 1970s. My findings support Minton's observations that these species are no longer common in those counties.

Species status.—I consider species of amphibians that were encountered in fewer than 5% of the sites that had the presence of at least one species, and species found in fewer than 10% of the counties, to be rare. By these criteria five species of frog and 12 species of salamander are considered rare. These species include the wood frog, eastern spadefoot toad, crawfish frog, pickerel frog, plains leopard frog, Jefferson salamander (*A. jeffersonianum*), northern slimy salamander, four-toed salamander (*Hemidactylium scutatum*), streamside salamander (*A. barbouri*), mudpuppy, longtail salamander, cave salamander, hellbender, red salamander, green salamander, ravine salamander, and northern dusky salamander.

I found additional information on the abundance of amphibians from surveys conducted in the 1990s and reported in the recent literature (Kolozsvary & Swihart 1999) and Indiana Department of Natural Resources reports. I combined additional amphibian survey data from 26 additional counties (Brown, Cass, Crawford, Dekalb, Elkhart, Harrison, Howard, Jackson, Jefferson, Jennings, Johnson, Kosciusko, Lagrange, Lake, La Porte, Marion, Marshall, Noble, Orange, Perry, Porter, Ripley, Saint Joseph's, Steuben, Tippecanoe, and Washington Counties) with my data to assess the recent abundance of each species in Indiana (Fig. 1). Some of this additional coverage, such as surveys of subterranean and river fauna of Blue River Watershed and areas in central and southeastern Indiana, fills gaps in my surveys.

Based on the larger data set (Table 5), the southern leopard frog, wood frog, pickerel frog, eastern spadefoot, crawfish frog and plains leopard frog are rare. Also note that each of these species was encountered much less often than two species that are on the state special concern list, the cricket frog (*Acris crepitans*) and northern leopard frog (*Rana pipiens*). In the larger data set, rare salamanders include the western lesser siren, marbled salamander (*Ambystoma opacum*), Jefferson salamanders, four-toed salamander, streamside salamander, mudpuppy, longtail salamanders, red salamander, hellbender, green salamander, ravine salamander-complex, and northern dusky salamander.

I consider species of reptiles that made up less than 5% of the 286 animals seen or captured, and found in less than 5% of the 156 sites that had at least one species of reptile, to be rare. Based on this criterion all reptile species are considered rare in Indiana with the exception of the painted turtle, common snapping turtle (*Chelydra serpentina*), eastern box turtle, six-lined racerunner, bullsnake, eastern

hognose snake, northern water snake, and common garter snake. I am more concerned about the possible declines of reptiles in Indiana than amphibians. However, this conclusion should be considered with caution because upland and riverine habitats were not targeted as rigorously as wetland habitats. Survey methods targeted for reptiles, such as drift-fence arrays, turtle traps and mark-recapture, should be employed in future surveys to establish a more robust and unbiased baseline data set.

The first stage of conservation planning is to review data on biodiversity of the region (Margules & Pressey 2000). After compiling data, the next step is to collect more information on the localities of species considered to be rare in the region. We need to distinguish between species that are threatened with extinction from those that were missed or underrepresented on the basis of habitats surveyed and methods used (Margules & Pressey 2000). Once these species-specific surveys are completed, then conservation goals, planning, and implementation of conservation actions should occur (Margules & Pressey 2000).

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CURRENT STATUS OF THE EVENING BAT, *NYCTICEIUS HUMERALIS*, IN INDIANA

John O. Whitaker, Jr. and **Sherry L. Gummer:** Department of Life Sciences, Indiana State University, Terre Haute, Indiana 47809 USA

ABSTRACT. As of 1993, we were not aware of any evening bat colonies still existing in Indiana. In 1994, evening bats were located in the Wabash River bottomlands of lower Prairie Creek, Vigo County, and later other colonies were located in the lower Wabash River Basin from Vigo to Posey Counties. Three additional colonies have also been discovered: one near Plainfield in Hendricks County, one near Zenas in Jennings County, and one in Bartholomew County at Camp Atterbury. Whereas all earlier colonies were in buildings, all known presently existing colonies are in trees. Evening bats probably lived in the bottomlands for many years and probably spread to upland buildings from there. The disappearance of evening bats from upland buildings may have been due to adverse pressure from the larger big brown bat, which has been increasing in the state. Eleven maternity colonies of evening bats had been known from Indiana and adjacent Illinois 30–40 years ago. All of these colonies were in buildings, and none of them exist today. One additional colony in a building was found in 1987 among 401 bat colonies in buildings in Indiana, but it too is now inactive. Data on bats from the Indiana State Department of Health for rabies examination also indicate a decline in this species in Indiana.

Keywords: Chiroptera, evening bat, bat, *Nycticeius humeralis*, status, Indiana

The evening bat, *Nycticeius humeralis*, is common in the southeastern United States, but relatively uncommon to the north. Seven records are included without comment from central and southwestern Ohio (Gottschang 1981), and Hoffmeister (1989) gives 31 specific records but lists it as rare throughout Ohio. The evening bat usually forms maternity colonies in buildings or in hollows in trees. It puts on a layer of fat and presumably hibernates, but almost nothing is known of this species in winter.

The evening bat was first recorded in Indiana by Charles M. Kirkpatrick in 1942 (Kirkpatrick 1943). Cope et al. (1961) collected information on 188 colonies of bats in buildings in Indiana by receiving calls in response to a widely-distributed poster asking for information. Four evening bat colonies in Clark, Clay, Orange, and Washington counties were located as a result of this survey (2.1% of the total), and information on distribution of bats of Indiana was summarized by Mumford & Cope (1964). They were then aware of six maternity colonies of this species in Indiana. In 1989 Whitaker and Gummer used the same survey procedure used by Cope et al. (1961) to locate bat colonies in buildings, but found only one evening bat colony among 401 bat colonies located (0.2%). This suggests that evening bats are decreasing in buildings in relation to other species. Humphrey & Cope (1970) reported on an additional colony in Montgomery County (from field notes of R.E. Mumford and J.B. Cope). Colonies were also known from Carroll, Cass, Clinton, Tippecanoe, and White counties in Indiana and Edgar County in Illinois, just west of the Indiana state line (Whitaker & Gummer 1993). Thus, colonies have been recorded in 11 counties in Indiana (Fig. 1) and adjacent Illinois as of 1993. Although all of these colonies were in buildings, all were close to larger streams flowing into the Wabash River in Indiana.

In addition to the 11 known colonies, there were five sites where earlier data (pregnant/lactating females or young taken) suggested nearby colonies. Information on the old sites is given by Whitaker & Gummer (1993). Whitaker & Gummer (1993) examined the roost sites and mist-netted in the vicinity of all the previous sites. Also James Mills, in 1986, mist-netted at some of these sites. No evidence of evening bats was found at any of the old sites.

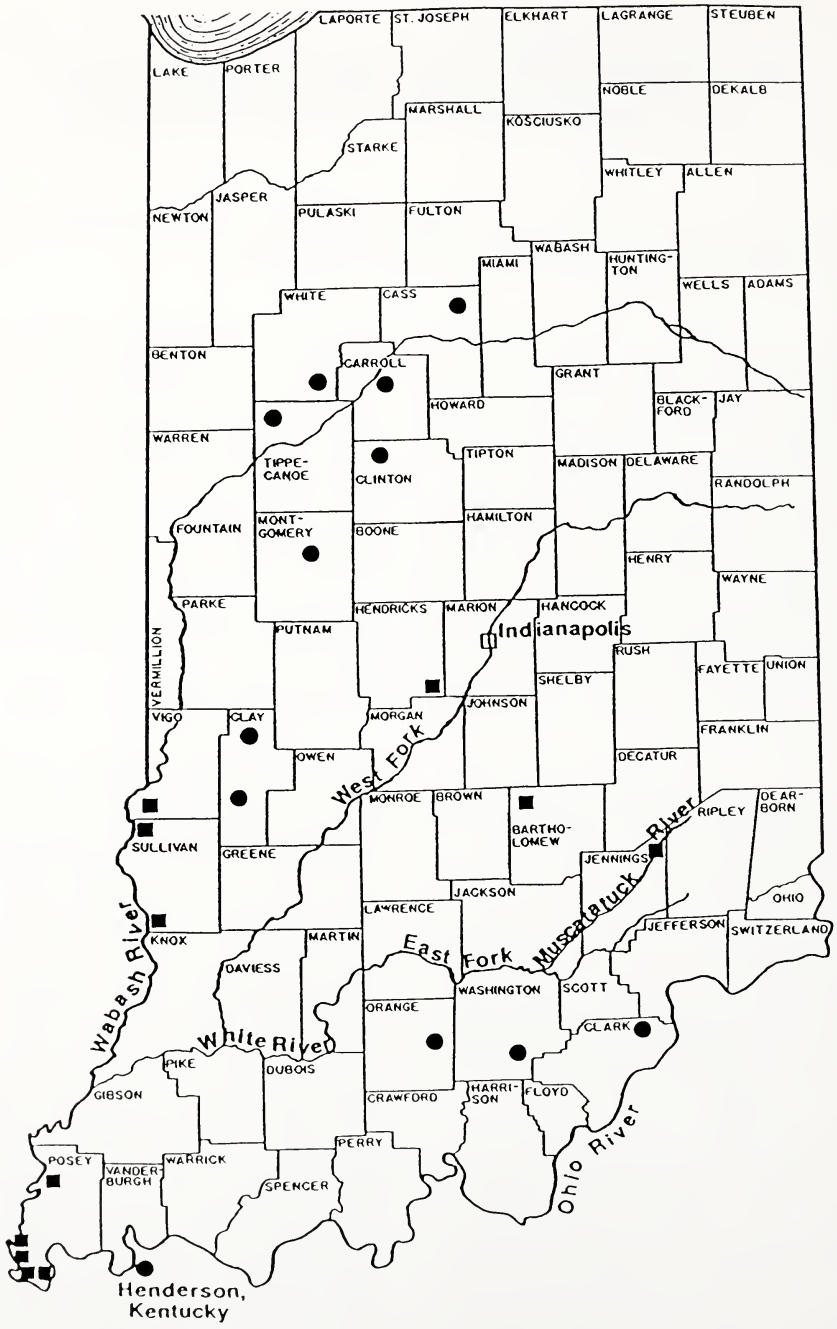

Figure 1.—Former and presently known colonies of the evening bat, *Nycticeius humeralis*, in Indiana. Circles (●) represent previously known, but now defunct, colonies. Squares (■) represent colonies known or presumed to be active in 2002.

Whitaker and Gummer distributed numerous posters in Indiana in order to locate evening bats. Of 401 colonies located during their studies, only one evening bat colony was found. It was located in a church, Briley Chapel, near Clay City, Clay County, Indiana, on 24 July 1987. However, this colony was evicted by church personnel in 1993. Much

information on evening bats from this colony was published by Clem (1992, 1993; Whitaker & Clem 1992; Whitaker et al. 1991). Thus we knew of no evening bats currently existing in Indiana as of 1993.

More recently we have learned much more about the distribution and behavior of evening bats in Indiana; the purpose of this paper is to provide an update on the current status of the evening bat in Indiana.

METHODS

A great deal of mist-netting has been done in Indiana in recent years by Whitaker and associates, and by Brack and associates (Whitaker et al. 2002; Whitaker & Gummer 2001). Included were 1067 nettings at sites in 80 counties. The timing and type of nettings varied with the project, but they occurred throughout the state. Most were with nets 5, 9, or 13 m wide and 2, 4, or 6 m high, and most were conducted from dusk until 0100 or 0200 h. One of these nettings (at Prairie Creek, Vigo County) resulted in the capture of six evening bats, leading to extensive netting at Prairie Creek. Consequently, weekly samplings of the area were initiated in 1994. From 1994 through 1999, we made 176 mist-nettings at Prairie Creek. Also, data from bats submitted to the Indiana State Department of Health rabies lab were included as well as preliminary data from two sites, Indianapolis International Airport (Hendricks County) and Camp Atterbury (Bartholomew County). Several evening bats were radio-tracked in late summer, 1994 at Prairie Creek (T10N R10W, Section 13, Hutton Quadrangle, 7.5′ USGS topographic map series).

RESULTS

During the 1067 nettings, we recorded 6445 individuals including all of the 10 species of bats currently existing in Indiana. On 25 July 1994, a site was sampled on lower Prairie Creek in Vigo County and 15 bats of six species were captured, including one Indiana myotis (federally-endangered) and six evening bats (state-endangered). These bats were taken in 650 ha of contiguous mixed bottomlands forest dominated in much of the area by silver maple, *Acer saccharinum*. The most abundant bat taken during the ensuing weekly nettings in the Prairie Creek area was the evening bat, *Nycticeius humeralis* (558 of 1439 bats taken, 3.17 bats per net-night), followed in order of decreasing abundance by the big brown bat, *Eptesicus fuscus* (254); the northern myotis, *Myotis septentrionalis* (237); the red bat, *Lasiurus borealis* (131); the little brown myotis, *Myotis lucifugus* (114); the eastern pipistrelle, *Pipistrellus subflavus* (92); the Indiana myotis, *Myotis sodalis* (49); the silver-haired bat, *Lasionycteris noctivagans* (3); and the hoary bat, *Lasiurus cinereus* (1). *Nycticeius humeralis*, and also *Eptesicus fuscus* and *Myotis septentrionalis* were taken at greater rates at Prairie Creek than in the lower Ohio or lower Wabash basin.

Several bats were radio-tagged at Prairie Creek. At first we expected to track them to buildings, as all the previously known evening bat colonies were in buildings. However, none of the radio-tagged bats roosted in buildings. Two were tracked to four separate hollow silver maple trees in the large bottomlands woods (Whitaker 1996). On 17 June 1995, a transmitter was placed on an adult female evening bat; and she was tracked to a silver maple tree. That night at least 350 bats emerged from what appeared to be a pileated woodpecker hole about 12 m up in that tree. More recently, S. Veilleux (unpubl. data) has tracked many more evening bats to a number of trees in these same woods.

Of the 6445 bats netted from throughout the state during 1811 nettings, 658 (10.2%) were evening bats (Table 1). However, although there was much netting throughout the state, most of the evening bats (558 individuals) were taken at Prairie Creek. Four other individuals from Vigo County were taken in a major tributary not far from the Wabash River. Almost all of the evening bats from Vigo County were juveniles or adult females. Some of the juvenile males in late summer had testes descended. In addition to the evening bats taken in Vigo County, 38 were taken in the lower Wabash Valley river bottom areas, 29 in Posey County, and nine in Sullivan County. Two from Sullivan County were females from Busseron Creek, thus indicating the presence of a maternity colony near there. The other seven evening bats were from just south of the Sullivan County line, undoubtedly from the Prairie Creek population. In Posey County, males were taken near Cypress Slough (Wabash Island Quad, T8S R4W, section 16), near Goose Pond (Wabash Island Quad, T7S

Table 1.—Bats caught in 1067 nettings from 80 of the 92 counties throughout Indiana (1987–1999).

	No. netted	No. bats/netting	No. counties	% of counties
Eptesicus fuscus	1748	1.63	68	85.0
Lasiurus borealis	1268	1.18	62	77.5
Myotis lucifugus	883	0.82	56	70.0
Pipistrellus subflavus	742	0.70	37	46.3
Myotis septentrionalis	741	0.69	36	45.0
Myotis sodalis	246	0.23	37	46.3
Nycticeius humeralis	658	0.62	12	15.0
Lasiurus cinerus	70	0.07	23	28.0
Lasionycteris noctivagans	16	0.01	11	13.8
Myotis grisescens	74	0.07	6	7.5
Total	6445			

R15W, Section 35), and at Hovey Lake (Uniontown Quad, T8S R14W, Section 14). Six of the 12 Posey County sites yielded a total of 29 evening bats. At one site an evening bat was radio-tagged and tracked to a silver maple tree across the Wabash River to the south in Illinois. Evening bats were also netted west of the Wabash River on the southern edge of Greathouse Island in a former bed of the Wabash River, on the Indiana/Illinois state line. Eighteen evening bats were netted there on 17 July 1996, including at least 13 females indicating that a maternity colony was very close to this site. It is clear that the evening bat is relatively common in the floodplain of the lower Wabash River in southwestern Indiana. We suspect that this species occurs commonly in the extensive bottomland woods in Gibson County, but this area has not been adequately netted. We have been unable to net it in Knox County, and suspect it is not common there because of a lack of bottomland woods.

Table 2.—Numbers of evening bats taken in various counties in Indiana since 1994, by mist-netting and by specimens submitted to the rabies laboratory, Indiana State Department of Health.

County	Mist-netting	Rabies laboratory
Vigo	562	—
Posey	29	3
Sullivan	29	—
Hendricks	36	—
Bartholomew	11	—
Vanderburgh	—	5
Jennings	5	—
Fayette	1	—
Floyd	—	1
Marion	—	1
Orange	1	—
Parke	1	—
Vermillion	1	—
White	1	—
	677	10

An even 600 of the 658 evening bats taken by recent mist-nettings by Brack and Whitaker (unpubl. data) were in the rather extensive river bottom woods of the southern Wabash River. Of the remaining evening bats taken, 36 were in Hendricks County, 11 were in Bartholomew County, 5 were in Jennings County, and 1 each were in Vermillion, Parke, Orange, Crawford, Fayette, and White counties (Table 2). Except for Fayette County, where one evening bat was taken, all of the records, new and old, are relatively near the Wabash or White rivers. Also, more recently many more evening bats have been netted at the Bartholomew County site (Camp Atterbury) and Hendricks County site (Indianapolis International Airport).

Bats submitted to the rabies laboratory of the Indiana Department of Health indicate the scarcity of this species. Only 38 of 7404 bats submitted to the lab between 1966–2000 were evening bats. These included 10 from 1966–1969, 12 from 1970–1979, 7 from 1980–1989, 8 from 1990–2000, and 4 in 2001. Three of these were from Posey County and 5 were from Vanderburgh County, in the lower Wabash area (Table 2). One from Marion County was in the White River drainage. The last was from Floyd County in the Ohio River basin.

DISCUSSION

Evening bat maternity colonies are presently known in at least five counties (Fig. 1). Evening bats have been netted in 11 counties since 1994 and 10 evening bats have been submitted to the Indiana state rabies laboratory from 1994–2001 (Table 2).

From the first netting, it was obvious that the Prairie Creek site contained an abundant and biologically diverse bat community and deserved intensive study. All of the pre-1993 *Nycticeius* maternity colonies in Indiana were associated with buildings (Mumford & Whitaker 1982), hence for some years we had been searching for this species in buildings. Therefore, the occurrence of relatively large numbers of evening bats in the bottomlands was of great interest. Whitaker & Gummer (1993) had previously noted that the colonies in Indiana were generally near the Wabash and White rivers, which is consistent with this idea. Numerous evening bats were netted in the Wabash River bottoms of Vigo to Posey counties. Prairie Creek and the Posey County sites, and to some extent the Sullivan County (Busseron Creek) site are large, flat, wooded bottomlands that are not developed or inhabited because they often flood. The abundance of evening bats in bottomland woods, where no netting had previously occurred, leads us to conclude that evening bats had been there historically and that the southern Wabash River bottomland woods are probably the ancestral habitat of this species.

Besides being along rivers, all evening bats radio-tracked since 1993 have roosted in trees, whereas all earlier known roosts in Indiana were in buildings. Roosts previously known from buildings may have been spillover from times when there were large populations in bottomland woods. Another possibility is that building roosts indicate that this species had adapted well to humans and their structures, similar to big brown bats and little brown myotis. However, the fact that the previously known populations in buildings are now all gone seems to indicate that this adaptation was not very successful, and perhaps that *Nycticeius* does not successfully compete with big brown bats for roosts in buildings, especially since the big brown bat has been increasing in Indiana (Whitaker, Brack & Cope 2002).

It is not known where evening bats hibernate. We have surmised that it could be in hollow trees near larger rivers to the south of Indiana or at least far enough south so that the temperature inside of the trees does not fall below freezing. Males are seldom associated with maternity colonies. All of the adult evening bats taken at Prairie Creek in 1999 (n = 145) and 2000 (n = 107) were females (total = 252), whereas 12 of 28 (42.9%) of the adult evening bats from Posey County were males. What is the reason then for the large number of males in Posey County? Perhaps the males remain to the south of the female range during the maternity season, and then migrate south with females. Another possibility is that southern Indiana is the northern edge of the hibernating range, and the males stay there all year. Additional data are needed on sex, age and temporal distribution of evening bats in Posey County to determine if the northern edge of the hibernating range for this species might be in southwestern Indiana.

It is clear that there are major populations of evening bats—at least in Vigo and Posey counties—where they roost in tree hollows (Fig. 1). Contrary to the situation in the lower Wabash Valley, no evening bats were netted in the Ohio River Valley east of Posey County (although one was taken in northern Kentucky, just east of Henderson, in 2002). This may be because there is relatively little bottomland forest in the Ohio Valley, at least on the Indiana side.

Earlier, there were colonies of evening bats in buildings north of Vigo County in north central Indiana, from Montgomery to Cass County (Fig. 1). We have netted very little in the bottomland woods of that area. It would be of interest to determine how far north the species extends in the Wabash River bottomlands.

In the Wabash River bottoms, this species lives in hollows in trees, where the bats commonly move between several trees, even when pups are present (unpubl. data). There is much movement between trees used as roosts by this species. Fairly large numbers of bats occur in some trees at times; at other times they are in smaller numbers. After the young become volant, movement between trees becomes greater. Since more trees are used, individual dusk counts are smaller.

We currently know of at least four other populations of evening bats in Indiana: (1)

near Zenas, in Jennings County, on the Muscatatuck River (Fig. 1). We radio-tracked a bat from this colony, but were unable to find the roost. We suspect that it was in a tree in a deep ravine there. (2) one on Honey Creek, south and east of Terre Haute in Vigo County. We tracked one bat from this colony to a tree in a yard, but a dusk count did not indicate that more bats were there, (3) southwestern Hendricks County, just south of the Indianapolis International Airport. Bats there were roosting in trees north of Interstate-70, but that woods was cut, and the evening bats moved south into a woodlot where a population of Indiana bats, *Myotis sodalis*, had resided for several years. The evening bats used several trees in this lot, possibly some of the same trees used by the Indiana myotis. (4) a population at Camp Atterbury in Bartholomew County. Many additional evening bats have been netted (and tracked) there.

Although evening bats commonly used buildings as roosts earlier, most now roost in trees. We know of none roosting in buildings at this time. It is possible that evening bats are being displaced in buildings by big brown bats, and are relatively successful in bottomland woods there are where few big brown bats. We suspect that trees in bottomland woods are the natural habitat for evening bats, and that they moved out from there into buildings, perhaps in good years when their population expanded.

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WINTERING POPULATIONS OF BATS IN INDIANA, WITH EMPHASIS ON THE ENDANGERED INDIANA MYOTIS, *MYOTIS SODALIS*

Virgil Brack, Jr.: Environmental Solutions & Innovations, LLC, 781 Neeb Road, Cincinnati, Ohio 45233 USA

Scott A. Johnson: Indiana Department of Natural Resources, 553 East Miller Drive, Bloomington, Indiana 47401 USA

R. Keith Dunlap: Indiana Karst Conservancy, P.O. Box 2401, Indianapolis, Indiana 46206 USA

ABSTRACT. Between 1981 and 2003, 269 visits were made to 72 caves in Indiana to count hibernating bats. Nearly 2,000,000 bats of seven species were counted: Indiana myotis (*Myotis sodalis*), little brown myotis (*M. lucifugus*), northern myotis (*M. septentrionalis*), gray myotis (*M. grisescens*), eastern pipistrelle (*Pipistrellus subflavus*), big brown bat (*Eptesicus fuscus*), and silver-haired bat (*Lasionycteris noctivagans*). Indiana myotis were found on 214 (80%) visits and in 30 (42%) caves. The statewide winter population of Indiana myotis found in 1981 (148,890) declined to a low of 99,202 in 1985 before reaching a record high of 185,899 bats in 1999. At least 50,000 Indiana myotis were found in Ray's Cave and in Twin Domes Cave; 10,000–50,000 each in Wyandotte, Jug Hole, Batwing, Coon, and Grotto caves; 1000–10,000 bats in Parker's Pit Cave; 100–1000 in each of 12 caves, and <100 bats in 10 other caves. The number of Indiana myotis in Twin Domes and Batwing caves, the largest populations in 1981, decreased >50% during the last 23 years. Declines in these two hibernacula were more than offset by gains at Rays, Wyandotte, Jug Hole, Coon, and Grotto caves. Little brown myotis were found during 226 (84%) visits in 49 (68%) caves; wintering populations in Indiana appear to be increasing. Big brown bats were found in 40 caves (56%) during 156 (58%) visits. The eastern pipistrelle was found in more caves (n = 61; 85%) and during more surveys (n = 234; 87%) than any other species. Northern myotis, gray myotis, and silver-haired bats were encountered infrequently.

Keywords: Bats, caves, hibernacula, Indiana, Indiana myotis, *Myotis sodalis*

While few species of bats from temperate regions use caves in summer (e.g., the gray myotis, *Myotis grisescens*, and Townsend's big-eared bat, *Corynorhinus townsendii*), use of caves as winter hibernacula is more common. Further, the winter range of several species, including the Indiana myotis (*Myotis sodalis*), is restricted to regions of well-developed limestone caverns or man-made mines. Most caves contain only a few hibernating bats but caves suitable for hibernation may contain large populations of several species.

Indiana has a significant cave resource (Powell 1966). Documented use of caves by bats is, however, often sporadic; and searches for new populations of bats are not a priority. Surveys are time consuming, difficult and sometimes dangerous, and there may be a misconception that known populations remain static over time.

Blatchley (1899) reported "tens of thousands" of little brown myotis, *Vespertilio subulatus*, (probably Indiana myotis) in Wyandotte Cave. Hall (1962) noted Indiana myotis in seven caves in southern Indiana, but considered only four (Wyandotte, Ray's, Coon, and Grotto) to contain significant populations. In 1976 and 1977, previously unknown concentrations of 100,000 and 50,000 Indiana myotis were found in two caves (Richter et al. 1978). Summarizing information from across the range of the Indiana bat, Humphrey (1978) reported 104,824 Indiana myotis in seven caves in Indiana, of which about 100,000 were in one cave. Humphrey & Cope (1976) reported wintering populations of little brown myotis (*Myotis lucifugus*) in 27 caves, 6 of which had sufficient numbers to warrant further studies. Mumford (1974) visited 12 caves

in or near the Hoosier National Forest in Harrison, Lawrence, Martin, Monroe, and Orange Counties. No Indiana myotis were found, but occasional occurrences of little brown myotis, big brown bats (*Eptesicus fuscus*), and eastern pipistrelles (*Pipistrellus subflavus*) were noted. Mumford (1974) also listed 19 known hibernacula of the Indiana myotis statewide, but numbers of bats were not provided. Mumford & Whitaker (1982) summarized known information, including use of caves by species in Indiana. However, their intent was an overview of species rather than detailed enumeration of caves surveyed and bats found.

During the winters of 1981–1983, Brack (1983) made 37 visits to 27 caves to document all species of bats encountered. Brack et al. (1984) summarized these and previous data for the Indiana myotis in the state. However, few data have since been provided to document this unique resource, although Whitaker & Brack (2002) detailed the distribution, including winter populations, of Indiana myotis in Indiana; and Whitaker et al. (2002) reported possible statewide declines of bats. Johnson et al. (2002) described cave management efforts for wintering colonies of Indiana myotis.

Our purpose is to report results of biennial surveys, over a 23-year period, of hibernacula in Indiana that contained known concentrations of Indiana myotis. We also include results of surveys to document new wintering populations of Indiana myotis and counts of winter concentrations of other cave-dwelling bats.

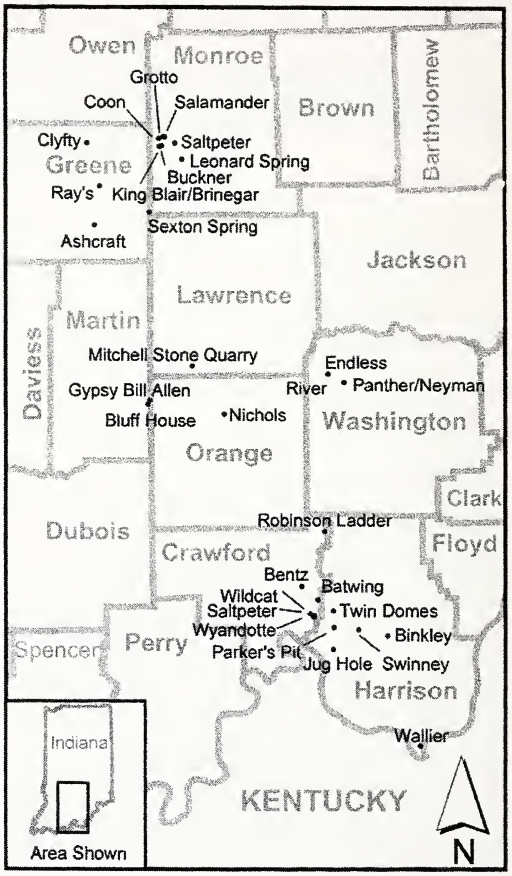


Figure 1.—Caves in Indiana in which populations of Indiana myotis were found during winter hibernacula surveys, 1981–2003.

METHODS

Between 1981 and 2003, 269 visits were made to 72 caves to count hibernating bats. To compare populations over time, data from 1981 and 1982 were composited to provide a more complete baseline, referred to as the 1981 estimate. Similarly, surveys in 1983 and 1985 were less complete than subsequent surveys, so statewide totals for Indiana myotis in those years were extrapolated for some smaller hibernacula. Extrapolations are included only in estimates of statewide populations. Numbers of bats in individual caves, and in individual or grouped surveys, do not include extrapolated numbers.

Caves surveyed were those known to have populations of Indiana myotis, reported by caving organizations to have concentrations of bats, or were likely to provide suitable habitat because of size, morphology, or airflow. Caves containing >100 Indiana myotis were visited every second year. Surveys were typically completed between mid-January and late February.

Bats were tallied by species and location in the cave. Individuals and small clusters of Indiana myotis were counted directly. When possible, larger clusters were measured with a tape measure or engineer's rule. Sizes of inaccessible clusters were initially estimated using visual-spatial associations, but laser calipers (i.e., two laser pointers bracketed to produce parallel beams 15.2 cm [6 inches] apart) were employed since 1997. Binoculars and a 1,250,000 candlepower spotlight (Collins Dynamics "Magnum" model search light) were used to count bats on high ceilings.

Cluster densities were usually estimated at 300 bats per 0.09 m^2 (1 ft^2; LaVal & LaVal 1980). When available, cave maps were used to locate clusters of bats and temperatures within the cave. Species of bats other than Indiana myotis were counted directly. Within caves, surveys were often terminated when few bats were encountered or warmer temperatures reduced the probability of encountering concentrations of bats.

Temperatures were taken outside entrances, in the twilight area, near Indiana myotis and concentrations of other species, and at intervals throughout the caves. Originally, a Schultheis quick-recording mercury thermometer was used exclusively. A Digi-Sense Model 8528-30 (J) thermocouple thermometer and 8116-40 general purpose probe were used for surveys in 1989 and 1991. Since 1991, most temperatures were taken using an infrared thermometer, speeding data collection and allowing temperatures to be obtained from inaccessible roost sites. Three Raytek models were used: ST2, Raynger® MiniTemp MT4, and Raynger® ST20.

RESULTS

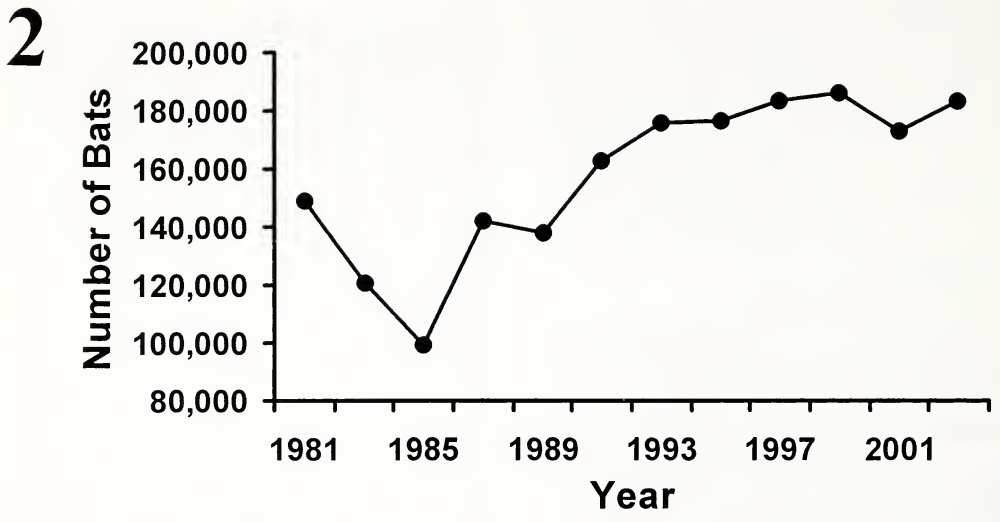
During this 23-year period, nearly 2,000,000 bats of seven species were found during 269 visits to 72 caves (Appendix 1). Caves were visited 1–13 times. The number of bats found per visit ranged from 0 to 98,250 ($\bar{x}$ = 7,314; *SD* = 16,973). Our surveys produced an average of 3.3 species (*SD* = 1.1) per cave. The number of caves visited, by county, was: Clark (*n* = 1), Crawford (*n* = 20), Greene (*n* = 4), Harrison (*n* = 9), Lawrence (*n* = 10), Martin (*n* = 4), Monroe (*n* = 10), Orange (*n* = 9), Owen (*n* = 1), and Washington (*n* = 4).

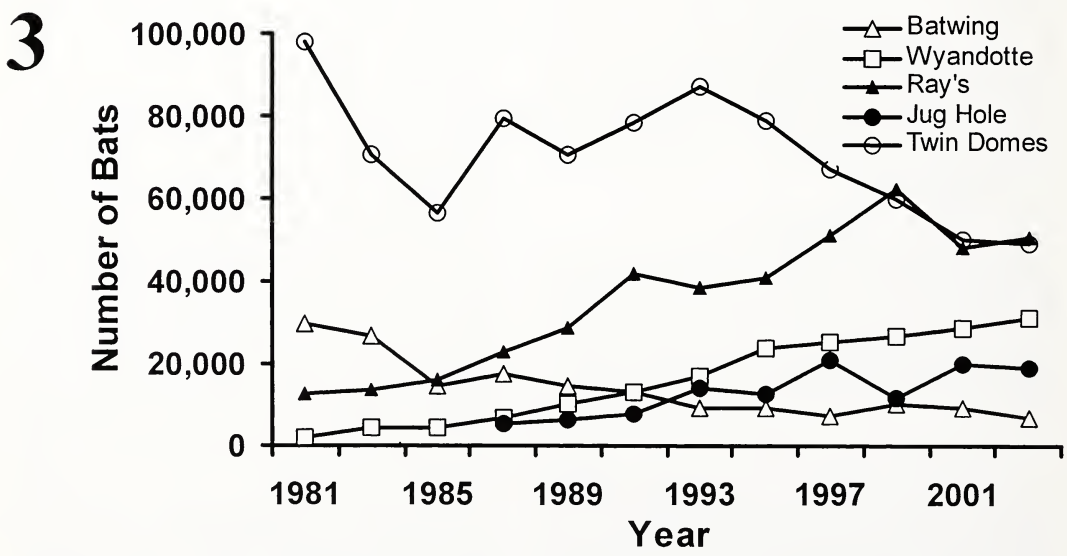
Indiana myotis.—Indiana myotis were found on 214 visits (80%) in 30 caves (42%; Fig. 1). Mean population size was 7081 bats (*SD* = 16,988), with the largest being 98,250 in Twin Domes Cave in 1981. At least 50,000 Indiana myotis were documented in Ray's Cave and in Twin Domes Cave; Wyandotte, Jug Hole, Batwing, Coon, and Grotto caves each contained 10,000–50,000 Indiana myotis; Parker's Pit Cave contained 1000–10,000 bats; and Buckner, Saltpeter (Crawford County), Saltpeter (Monroe County), Clyfty, Endless, Leonard Springs, Robinson Ladder, Sexton Springs, Wallier, King Blair/Brinegar, Panther/Neyman, and Gypsy Bill Allen caves each contained 100–1000 bats. Ten caves each had <100 Indiana myotis: Wildcat, River, Salamander, Ashcraft, Bentz, Mitchell Crushed Stone Quarry, Binkley, Swinney, Nichols, and Bluff House caves.

The number of Indiana myotis hibernating in the state has varied over time. In 1981, the known population was 148,890 (Fig. 2). By 1985, the total had decreased 33% to a low of 99,202 bats. Statewide winter populations in Indiana increased through 1999 to a high of 185,899 bats. Increases at Ray's and Wyandotte caves were largely responsible for the observed increase; but newly discovered hibernacula, most notably Jug Hole Cave (19,240 bats in 2003), contributed to the increase. The 2001 survey, which tallied 173,076 Indiana myotis, was the smallest total in nearly 10 years. The 2003 survey tallied 183,298 bats (Fig. 2).

Numbers of Indiana myotis using specific hibernacula have changed over time (Fig. 3). In 1981, there were two known major hibernacula in Indiana, Twin Domes and Batwing caves, which collectively held an estimated 128,210 Indiana myotis, 86% of the statewide total. Ray's Cave contained the next largest population, 12,500 bats, and Wyandotte Cave, with 2152 Indiana myotis, the fourth largest population. Jug Hole Cave, first surveyed in 1987, held an estimated 5535 Indiana myotis. Between 1981 and 2003, the combined estimated population of Twin Domes and Batwing caves decreased to 56,250 bats, 31% of statewide total. In 2003, Ray's, Wyandotte, and Jug Hole caves collectively held 101,398 bats, 55% of statewide total.

In 1981, small populations of Indiana myotis were known from 11 other caves, with a collective population of only 6028 bats ($\bar{x}$ = 553; *SD* = 942), or 4% of the population of Indiana myotis in the state. This included 1190 bats in Coon and 3190 bats in Grotto. Between 1981 and 2003, 14 additional caves with small populations of bats were documented. By 2003, we found 10,675 bats in Coon Cave and 10,338 bats in Grotto Cave; and the collective populations in 22 of 25 small hibernacula contained 25,650 Indiana myotis ($\bar{x}$ = 1165; *SD* = 3033), or 14% of the state's total. Zero to three Indiana myotis were found in the most recent visits of the remaining three hibernacula.





Figures 2, 3.—Numbers of Indiana myotis observed during winter hibernacula surveys in Indiana, 1981–2003 in all hibernacula (Fig. 2), and in each of the five largest hibernacula (Fig. 3).

In addition to changes in wintering populations in specific caves, portions of hibernacula used by Indiana myotis have changed over time. To illustrate, in Wyandotte Cave, three areas, each progressively deeper in the cave (i.e., warmer) received the most use: entrance, Washington Avenue, and Rugged Mountain. Through 2001, from <1% to 7.1% used the entrance area, 68.8–87.0% used Washington Avenue, and 7.6–25.1% used Rugged Mountain. Between 1995 and 2001, numbers of Indiana myotis in Rugged Mountain increased from an estimated 1897 (7.9%) to 7167 bats (25.1%). During the 2003 survey, the first since winter cave tours were discontinued, 4368 bats (14.0%) hibernated in Bats Lodge, a portion of the cave unused during the preceding 22 years. Bats Lodge is an area with a low ceiling through which tours passed. The use of Bats Lodge was associated with a decrease in the number of bats in Washington Avenue, where 19,087 bats, or only 61.1% of the population, was found. The proportions of bats in the entrance area (3.1%; 962 bats) and

Rugged Mountain (21.0%; 6570 bats) were within the range of past use.

Little brown myotis.—Little brown myotis were found during 226 (84%) visits ($\bar{x}$ = 237; *SD* = 420) in 49 (68%) caves, and in every county where surveys were completed except Clark (Appendix 1). The largest population was 3380 in Ray's Cave in 1981. Concentrations ≥1000 little brown myotis were found only in Ray's Cave ($\bar{x}$ = 1051; *SD* = 928), Grotto Cave ($\bar{x}$ = 962; *SD* = 800), and Endless Cave ($\bar{x}$ = 750; *SD* = 435). From 500–1000 little brown myotis were found in Wildcat ($\bar{x}$ = 388; *SD* = 103), Parker's Pit ($\bar{x}$ = 234; *SD* = 174), Coon ($\bar{x}$ = 277; *SD* = 186), River ($\bar{x}$ = 301; *SD* = 137), Batwing ($\bar{x}$ = 181; *SD* = 254), and King Blair/Brinegar ($\bar{x}$ = 353; *SD* = 159) caves. Eight additional caves contained ≥250 little brown myotis.

Numbers of little brown myotis counted ranged from 1406 in 1985 to 8025 in 2001 (Fig. 4), while the mean number of bats per cave ranged from 106 (1989) to 968 (1983). Both totals and averages suggest wintering populations have been increasing since the mid- to late 1980's, concurrent with increases of the Indiana myotis. Consistent increases were noted at Coon, Grotto, and Endless caves, and erratic increases were noted at Saltpeter (Crawford County) and Parkers Pit caves. In contrast, numbers of little brown myotis in Ray's and Panther/Neyman caves decreased and an erratic decrease occurred in King Blair/Brinegar Cave.

Northern myotis.—Northern myotis (*Myotis septentrionalis*) were widespread but found in low numbers. Usually only one to several individuals were found during any year of cave visits. Occurrence was not restricted to any cave or region of the state. For example, northern myotis were found more frequently in 2003 than in most winters, and 25 individuals were found in 11 of 28 caves visited. Ten individuals, a record high, were found in Endless Cave. Bats of this species hibernated individually, often in cracks or other tight spots, and may be overlooked.

Gray myotis.—Gray myotis were rarely found. One or two gray myotis were found on four surveys in Twin Domes and Wyandotte caves, at the northern periphery of this species range. Most recently, a single gray myotis was found in Twin Domes Cave in 2003.

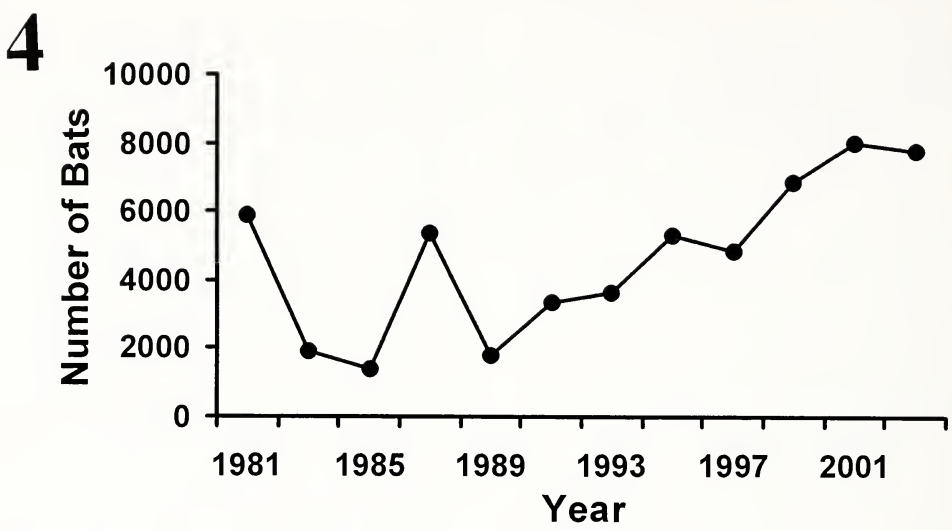
Big brown bat.—Big brown bats were found in 40 caves (56%) during 156 (58%) visits (Appendix 1). They were found in every county where surveys were completed except Clark. Mean number per cave was 11 bats (*SD* = 26). The largest concentration was 224 in Mitchell Crushed Stone Quarry Cave in 1991. One additional cave, Ray's, has held at least 100 big brown bats (1993 and 1995). In 1995, 53 big brown bats hibernated in Wyandotte Cave, the only hibernaculum and only survey in which 50–100 individuals were found. Neither annual totals (134–431), nor mean number of bats per cave (8–55), suggest a change in numbers of big brown bats hibernating in caves of Indiana (Fig. 5). Further, no individual cave has an increasing or decreasing population of big brown bats.

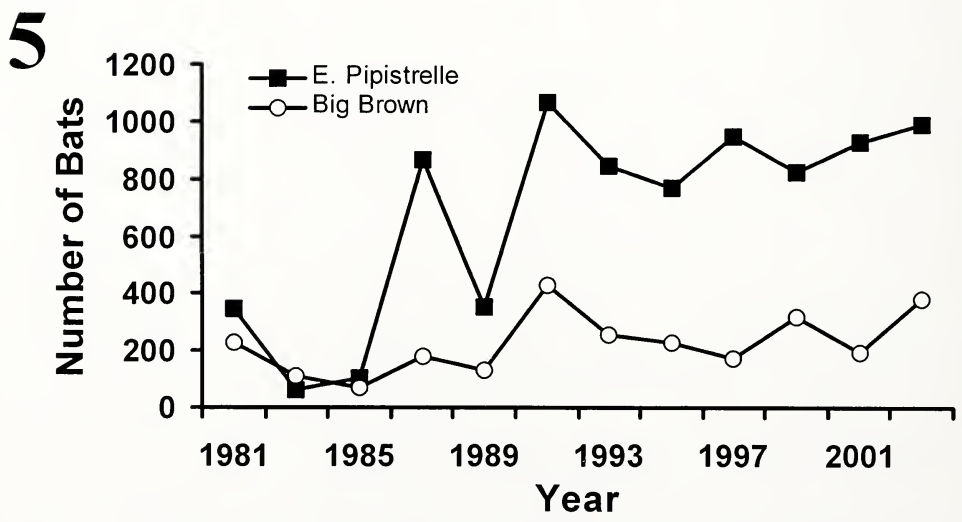
Eastern pipistrelle.—The eastern pipistrelle, the most common species, was found in 61 caves (85%) during 234 (87%) visits (Appendix 1). Numbers of bats per cave were variable, as were numbers of bats per visit ($\bar{x}$ = 35; *SD* = 48). The largest number (278) was found in Coon Cave in 1991. Populations >200 eastern pipistrelles were also found in Clyfty (1997) and King Blair/Brinegar (1987 and 1995) caves. River Cave had 153 eastern pipistrelles in 1991 and Grotto Cave had 100 in 1997. Thirteen additional caves contained at least 50 individuals during one or more surveys. Neither annual totals (62–1071) nor mean number of bats per cave (15–54) indicate that numbers of eastern pipistrelles hibernating in Indiana caves are increasing or decreasing (Fig. 5). Similarly, there are no individual caves showing a consistent change over time. Indeed, numbers of eastern pipistrelles in many caves varied widely among visits.

Silver-haired bat.—The silver-haired bat (*Lasionycteris noctivagans*) was the species least frequently encountered during cave visits. One individual was found hibernating in Wyandotte Cave in 1989, and two individuals were found in approximately the same location in 1991.

DISCUSSION

The Indiana myotis has been known from hibernacula of the state for over 100 years. Because the species was listed as endangered by the U.S. Fish and Wildlife Service, documentation of this resource, numbers of bats and location of hibernacula, and changes over time, are important for management of the





Figures 4, 5.—Numbers of little brown myotis (Fig. 4), and eastern pipistrelle and big brown bats (Fig. 5) observed during winter hibernacula surveys in Indiana, 1981–2003.

species. Measures to prevent human disturbance, maintain a suitable thermal regime, and forestall natural disasters can be implemented (Johnson et al. 2002).

Increases of Indiana myotis were most notable at hibernacula that had smaller populations both historically and when our surveys began in 1981 (e.g., Ray's, Wyandotte, Coon, and Grotto caves). Conversely, caves that contained the largest concentrations 23 years ago (i.e., Twin Domes and Batwing caves) have sustained large declines. In addition, hibernacula unknown in the early 1980's, such as Jug Hole, Leonard Spring, Robinson Ladder, Sexton Spring, Wallier, and Gypsy Bill Allen caves, were found and in some cases, particularly Jug Hole Cave, experienced significant increases. Finally, caves such as Endless and King Blair/Brinegar, which did not previously contain Indiana myotis, now support important populations. Statewide, increases have more than offset losses and inter-cave movements do not explain all changes, which indicates a true increase in the numbers of Indiana myotis hibernating in Indiana during the last 23 years.

Changes in hibernacula use, for whatever reasons, question our knowledge of hibernacula suitability for this species. Management standards are based on limited data obtained 25 years ago from a few hibernacula, many of which have since experienced marked declines. In many cases, changes were unrelated to disturbance: protected hibernacula experienced the greatest losses while some hibernacula gaining bats had little or no protection (Johnson et al. 2002). Documenting the physical environment, most notably hibernacula temperatures, may help elucidate reasons for shifts in use. Our understanding of needs of the species and development of management practices may be based on initial observations of populations in suboptimal conditions.

Changes in populations among hibernacula and intra-cave variability in hibernation sites suggest the species is adaptable and seeks roost sites that provide specific conditions for winter hibernation. For example, numbers and locations of bats in portions of Wyandotte Cave varied since bats were first reported from the cave. Blatchley (1899) reported "tens of thousands" *Vespertilio subulatus*, (i.e., Indiana myotis) in Odd Fellows' Hall. Since then, this cave has endured extensive alterations. The entrance was enlarged during commercialization of the cave early in the 1950's, and a rock wall was placed across the entrance. This wall was replaced in 1978 with a partial wall and a flat-iron gate, and again in 1991 with an angle-iron bat gate. Many areas were dug out to explore and commercialize the cave. One large alteration was 'opening' of the new section in 1941, which undoubtedly altered airflow. No Indiana myotis have been found in the Odd Fellows' Hall area since opening of the new section.

In 1955, only 500 Indiana myotis were estimated to hibernate in Wyandotte Cave, all in the Washington Avenue area. In 1981, 2151 Indiana myotis were tallied in the cave (Brack et al. 1984), and the population has steadily grown to 31,217 individuals in 2003. Further, the location and size of clusters in Washington Avenue varied among surveys; bats also now hibernate in the entrance, Rugged Mountain, and Bats Lodge areas (Whitaker et al. 2003). Use of Rugged Mountain appears to be increasing, and substantial use of Bats Lodge was first documented in 2003.

Only about 500 Indiana myotis used Ray's Cave in 1960 (Hall 1962), and 2700 were found in 1975 (Brack et al. 1984). In 1981, Ray's Cave harbored 12,500 bats, and the population increased to a recorded high of 62,464 in 1999. The most recent survey, in 2003, totaled 50,941 individuals.

Only nine Indiana myotis were in Coon Cave in 1960 (Hall 1962), and 24 were found in 1975 (Humphrey 1978). In 1982, 550 Indiana myotis were found (Brack et al. 1984), but by 2003, the population was tallied at 10,675 individuals. Similarly, numbers of Indiana myotis using Grotto Cave were about 200 in 1960 and 1975, while the 2003 survey produced 10,338 bats. In contrast, numbers of Indiana myotis in nearby Buckner's Cave decreased from 345 in 1975 (Humphrey 1978) to a single individual in 2001. Much of this decline was attributable to human disturbance. An increase to 40 Indiana myotis in 2003 may be attributable to reduced disturbance.

Richter et al. (1978) reported >100,000 Indiana myotis in Twin Domes Cave in 1976 and 50,000 bats (±20%) in Batwing Cave in 1977. Populations in both caves have steadily declined. In 2003, there were 49,350 Indiana myotis in Twin Domes Cave, about a 50% decline since the population was discovered. Only 6900 Indiana myotis were found in Batwing Cave in 2003, an 86% decline.

Our data suggest wintering populations of little brown myotis have increased in the past 20 years. In contrast, Whitaker et al. (2002) presented evidence, based largely on summer occurrences, of a decline of this species in the state. Numbers of hibernating little brown myotis increased during the same period when the Indiana myotis increased. Perhaps similar factors, such as less disturbance in hibernacula or elements in summer habitats, affected both species similarly. Because our surveys emphasized Indiana myotis, some little brown myotis were likely not found. However, concentrations of little brown myotis, like Indiana myotis, are generally found in cold, anterior portions of caves, where survey was likely. Nevertheless, individuals and small numbers of little brown myotis are found in almost any part of a cave, including warmer areas.

Similarly, our emphasis on Indiana myotis likely underestimated wintering populations of eastern pipistrelles and big brown bats. The eastern pipistrelle, often found in warmer caves and in parts of caves that do not cool

sufficiently for other species, is more widely distributed than Indiana myotis. Big brown bats are often found near cave entrances, but they sometimes hibernate deeper within the cave. Big brown bats frequently hibernate in houses and outbuildings (Whitaker 1997; Whitaker & Gummer 1992, 2000), further limiting interpretation of wintering populations in caves. Our data do not indicate any appreciable change in numbers of big brown bats and eastern pipistrelles hibernating in Indiana caves. Whitaker et al. (2002) presented evidence, again based largely on summer occurrences, of increases of both species in the state.

It is likely that most caves in Indiana that support relatively large concentrations of bats have been found in the last 23 years. Nevertheless, efforts should continue to monitor changes in existing populations and to identify new hibernacula. Populations that are currently insignificant may increase, and new populations may become established. For example, nearly 10,000 Indiana myotis and 20,000 little brown myotis were found in 1995 in a limestone mine in southwest Ohio (VB unpubl. data). This mine ceased operation only about 15 years before the bats were found.

ACKNOWLEDGMENTS

Many individuals assisted with field studies, and any attempt to list them would be incomplete. We simply state our sincere thanks to all of them. Individuals from conservation and caving organizations identified concentrations of bats or potentially suitable caves for further investigation. Again, we express our thanks. The Indiana Division of Fish and Wildlife funded most field studies, and we thank Catherine Gremillion-Smith for support. The Nature Conservancy and U.S. Forest Service, North Central Forest Experiment Station provided funds for surveys 1981–1983. Hoosier National Forest funded 2002 field studies; we thank Kelle Reynolds for support. Richard Clawson, Missouri Department of Conservation, provided data on Batwing and Twin Domes Caves, collected on behalf of the U.S. Fish and Wildlife Service. Two anonymous reviewers made helpful comments on an earlier draft of the manuscript.

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Appendix 1.—Numbers of bats of four species observed during winter hibernacula surveys in Indiana, 1981–2003.

Cave	Year	*Myotis sodalis*	*Myotis lucifugus*	*Eptesicus fuscus*	*Pipistrellus subflavus*
Clark County					
Indian	1985	0	0	0	0
Crawford County					
Across The Valley	2002	0	0	0	2
Batwing	1981	29,960	1	0	11
	1983	26,650			
	1985	14,750			
	1987	17,450			
	1989	14,500			
	1991	13,150			
	1993	9,350			
	1995	9,300			
	1997	7,400			
	1999	10,125			
	2001	9,350			
	2003	6,900	360	0	16
B-B Hole	1999	0	156	0	9
Bentz	1982	0	16	8	24
	1989	3	86	2	32
	2001	0	124	2	56
Big Circle	2002	0	0	0	5
Big Windy (Bauer's Blowhole)	1997	0	24	0	1
Easter Pit	1995	0	108	0	7
Hermit	2002	0	2	0	0
Little Jug Hole	1995	0	1	0	7
Little Mellet Pit	1989	0	0	0	0
Mellet Pit	1989	0	0	0	1
Mesmore Springs	2002	0	112	1	91
Pavey	2002	0	1	1	3
Robinson Ladder	1989	95	14	0	7
	1991	388	8	0	26
	1993	376	8	0	28
	1995	219	182	0	3
	1997	326	0	0	0
	1999	223	119	0	0
	2001	366	58	2	2
	2003	204	164	0	0
Saltpeter	1982	352	114	8	7

Appendix 1.—Continued.

Cave	Year	*Myotis sodalis*	*Myotis lucifugus*	*Eptesicus fuscus*	*Pipistrellus subflavus*
	1987	427	198	7	25
	1989	295	28	0	7
	1991	508	154	12	60
	1993	375	76	7	15
	1995	647	182	7	39
	1997	577	302	11	68
	1999	800	204	4	10
	2001	849	301	1	29
	2003	681	250	3	32
Sentinel Rock	2002	0	0	0	0
Treasure	2002	0	0	0	0
Westerhoff	1989	0	0	0	3
Wildcat	1982	29	332	0	30
	1987	0	520	0	63
	1991	31	310	0	33
	1993	61	314	0	19
	1995	34	582	0	45
	1997	48	359	0	49
	1999	19	344	0	18
	2001	35	446	0	28
	2003	17	289	0	13
Wyandotte	1981	2,152	6	11	2
	1983	4,550			
	1985	4,627	21	2	1
	1987	6,681	275	12	2
	1989	10,344	8	32	14
	1991	12,994	15	11	21
	1993	17,304	12	38	4
	1995	23,878	30	53	18
	1997	25,424	70	24	32
	1999	26,854	268	17	13
	2001	28,584	272	10	16
	2003	31,217	15	14	16
Greene County					
Ashcraft	1993	20	190	4	6
	1995	28	170	0	14
	1999	3	29	0	0
Clyfty	1982	66	298	10	46
	1987	198	295	17	124
	1989	412	233	9	73
	1991	357	334	15	106
	1993	307	176	1	53
	1995	299	141	1	93
	1997	369	268	6	212
	1999	379	272	2	87
	2001	469	394	26	104
	2003	457	329	26	139
Ray's	1981	12,500	3,380	60	14
	1982	11,822	799	95	10
	1983	13,475	1,834	85	14
	1985	16,200	1,044	59	15
	1987	22,990	2,395	74	38
	1989	28,581	671	53	10

Appendix 1.—Continued.

Cave	Year	*Myotis sodalis*	*Myotis lucifugus*	*Eptesicus fuscus*	*Pipistrellus subflavus*
	1991	41,854	600	88	94
	1993	38,386	351	118	33
	1995	41,157	677	108	84
	1997	51,365	316	37	58
	1999	62,464	686	59	43
	2001	48,219	644	41	70
	2003	50,941	267	31	53
Sexton Spring	1991		1	4	1
	1993	67	196	4	89
	1995	117	64	0	3
	1997	98	7	0	0
	1999	75	17	0	1
	2001	100	87	0	2
	2003	113	21	3	0
Harrison County					
Binkley	1997	84	197	0	17
	2001	9	110	0	25
Borden's Pit	1991	0	13	0	26
Harmon Pit	2003	0	121	1	62
Jug Hole	1987	5,535	9	0	6
	1989	6,424	5	13	9
	1991	7,640	15	16	12
	1993	13,924	9	10	3
	1995	12,463	8	6	10
	1997	20,741	12	11	17
	1999	11,900	10	2	12
	2001	20,151	11	0	8
	2003	19,240	12	9	9
Parker's Pit	1982	500			
	1987	1,803	101	10	18
	1989	1,104	141	5	6
	1991	926	110	9	14
	1993	1,045	209	4	7
	1995	1,276	406	4	4
	1997	1,139	234	2	6
	1999	987	106	5	5
	2001	989	622	3	4
	2003	447	181	3	5
Penny Well	1995	0	0	0	2
Swinney	1997	11	0	0	0
	1999	29	3	13	1
	2001	39	2	5	1
	2003	184	0	7	0
Twin Domes	1981	98,250	0	0	0
	1983	70,750			
	1985	56,650			
	1987	79,650			
	1989	70,800			
	1991	78,500			
	1993	87,350			
	1995	78,875			
	1997	67,100			
	1999	59,775			

Appendix 1.—Continued.

Cave	Year	*Myotis sodalis*	*Myotis lucifugus*	*Eptesicus fuscus*	*Pipistrellus subflavus*
	2001	50,325			
	2003	49,350	1	0	4
Wallier	1991	36	7	23	7
	1993	72	1	25	18
	1995	465	1	15	9
	1997	409	3	48	18
	1999	381	4	7	6
	2001	310	9	4	8
	2003	541	8	24	19
Lawrence County					
Bronson's	1982	0	0	0	3
Dixon Pit	1993	0	133	5	75
Doghill-Donnehue	1982	0	98	6	2
	1989	0	119	11	14
Donaldson's	1982	0	0	25	3
Hamer	1982	0	1	10	1
King	1987	0	0	0	0
	1989	0	0	4	0
Mitchell Crushed Stone Quarry	1991	9	178	224	41
	1999	31	380	162	63
	2003	38	280	175	65
Salts	1982	0	33	6	12
	1993	0	102	25	48
Sullivan	1982	0	0	0	0
Twin	1982	0	0	1	0
Martin County					
Aunt Liz	1989	0	1	0	41
Bluff House	2002	1	0	0	7
	2003	0	2	2	4
Granny's	1989	0	1	0	14
Gypsy Bill Allen	2001	134	44	15	34
	2003	250	24	11	21
Monroe County					
Brinegar	1987	12	363	0	205
King Blair/Brinegar	1993	442	427	0	101
	1995	514	673	0	275
	1997	663	295	0	106
	1999	453	247	0	72
	2001	263	220	0	57
	2003	190	248	0	89
Buckner	1982	488	32	2	27
	1985	301	21	9	0
	1987	336	29	0	12
	1989	24	16	0	9
	1991	51	16	0	9
	1993	25	23	2	3
	1995	41	35	3	8
	1997	15	15	2	8
	1999	6	41	2	10
	2001	1	54	0	6
	2003	40	46	4	5
Coon	1981	1,190	31	0	6

Appendix 1.—Continued.

Cave	Year	*Myotis sodalis*	*Myotis lucifugus*	*Eptesicus fuscus*	*Pipistrellus subflavus*
	1982	550	12	1	5
	1985	777	20	2	5
	1987	2950	152	3	166
	1989	2103	176	5	103
	1991	3696	394	4	278
	1993	4451	392	7	208
	1995	4455	423	1	200
	1997	4786	380	3	188
	1999	6341	376	2	189
	2001	6395	443	0	147
	2003	10,675	525	2	135
Eller's	1987	0	0	0	3
Grotto	1981	3,190	589	0	2
	1982	2,692	1,090	0	44
	1985	4,198	291	0	8
	1987	3,778	311	0	1
	1989	2,985	213	0	0
	1991	1,996	178	1	5
	1993	1,568	338	2	8
	1995	2,018	937	4	32
	1997	2,435	1,143	5	44
	1999	4,361	1,811	9	100
	2001	5,419	2,276	1	57
	2003	10,338	2,363	1	85
Leonard Spring	1989	135	94	0	17
	1991	112	176	0	68
	1993	92	121	2	81
	1995	82	260	0	96
	1997	92	195	0	35
	1999	81	198	0	61
	2001	25	231	0	48
	2003	138	164	1	38
Patton	2002	0	1	4	3
Ranard School	1982	0	0	8	0
Salamander	1982	74	130	0	21
	1987	0	138	1	79
	1991	0	124	0	50
	1997	1	8	0	0
	2003	0	19	0	4
Saltpeter	1982	83	19	46	0
	1987	19	0	33	1
	1991	221	68	14	12
	1993	245	79	16	20
	1995	175	65	21	17
	1997	136	38	5	5
	1999	40	29	4	9
	2001	24	41	8	15
	2003	96	51	14	15
Orange County					
Diggers Delight	2002	0	0	0	3
Dillon	2002	0	2	1	51
Duggins Springs	2002	0	0	0	6
Elrod	2001	0	0	2	93

Appendix 1.—Continued.

Cave	Year	*Myotis sodalis*	*Myotis lucifugus*	*Eptesicus fuscus*	*Pipistrellus subflavus*
	2002	0	0	0	98
Nichols	2003	39	26	4	18
Red Berry	2002	0	16	0	6
Spring Springs	2002	0	0	0	7
Tucker Lake Springs	2002	0	0	3	8
Wesley Chapel Gulf	2002	0	1	0	7
Owen County					
Boone's	1987	0	1	10	18
	2001	0	4	20	24
Washington County					
Endless	1982	2	163	17	26
	1987	1	330	11	29
	1991	134	460	9	55
	1993	335	602	9	74
	1995	450	772	9	74
	1997	404	665	10	68
	1999	403	993	25	44
	2001	800	1,486	46	89
	2003	863	1,275	33	60
Mill	1985	0	9	0	74
Panther/Neyman	1995	86	277	0	5
	1997	156	249	7	10
	1999	167	246	3	5
	2001	220	146	5	5
	2003	337	239	15	5
River	1982	104	170	4	65
	1987	5	242	2	75
	1991	1	216	1	153
	1999	2	368	2	65
	2003	2	509	4	81

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BATS OF WYANDOTTE CAVE, CRAWFORD COUNTY, INDIANA

John O. Whitaker, Jr.: Department of Life Sciences, Indiana State University, Terre Haute, Indiana 47809 USA

James B. Cope[1]**:** Department of Biology, Earlham College, Richmond, Indiana 47374 USA

Virgil Brack, Jr.: Environmental Solutions and Innovations, 781 Neeb Road, Cincinnati, Ohio 45233 USA

ABSTRACT. Nine of the 12 species of bats known from Indiana have been observed at Wyandotte Cave during mist-netting or trapping at the entrance and by in-cave counts from 1953 to the present. The most abundant bat at Wyandotte Cave is the Indiana myotis, *Myotis sodalis*, and the numbers of bats of that species hibernating in the cave have been increasing steadily over the last two decades. The second most abundant bat is the little brown myotis, *Myotis lucifugus*. At least prior to 1980, little brown myotis outnumbered Indiana myotis during swarming; but that has changed as the hibernating population of Indiana myotis increased. Relatively few eastern pipistrelles (*Pipistrellus subflavus*) or big brown bats (*Eptesicus fuscus*), are found at Wyandotte Cave, and *Myotis septentrionalis* is even less common. One southeastern myotis (*Myotis austroriparius*, now extirpated in Indiana), was seen in hibernation there in February 1966. One gray bat (*Myotis grisescens*) was found in hibernation in the cave in February 1967, and several have been netted at the entrance. Also, a few male gray bats were found inside the cave in summer forming a small bachelor colony. Two silver-haired bats, *Lasionycteris noctivagans*, were found in hibernation; and two were captured by mist-netting, both in late March. Red bats, *Lasiurus borealis*, are seldom found in caves, but 43 were captured by mist-netting at the entrance.

Keywords: Bats, Chiroptera, hibernation, Wyandotte Cave

Wyandotte Cave has been known since 1798. It is in Crawford County, Indiana, and is located in Harrison-Crawford State Forest. It has 14.8 km of known mapped passages (Fig. 1). Over the years, it has been used as a saltpeter-epsom salt mine, domestic and commercial produce warehouse; and it has long been used as a commercial cave for tourists. The cave has a large, downward sloping entrance and currently has a bat-friendly gate, constructed in 1991, about 100 m into the cave. It has been owned and operated by the Indiana Department of Natural Resources since 1966. It was visited by many people each year, including tours in winter. Johnson et al. (1998) recorded more than 375 tours between mid-September and late April in each of two years. However, the cave has been privately operated since 2002, and winter tours have been terminated.

Henry P. Rothrock apparently purchased the property containing the cave in 1820 from the U.S. Land Office in Jeffersonville, Indiana. The first good descriptions of the cave entrance were made by Adams (1818) and McMurtrie (1819), after almost a decade of mining. The entrance was about 4–5 m wide, and about 1–1.5 m in height, descending by gradual steps into a large, spacious room. In the first few years a stone wall with a small wooden door was built a few meters inside the drip line. A portion of this wall is still present to the right of the trail leading into the cave. This wall had the effect of shortening the horizontal opening from 4–5 m to perhaps 2.2 m. The cave remained semi-developed for the first 50–70 years as a commercial cave, but few improvements were made since Rothrock desired to keep the cave pristine.

Doors and gates can greatly affect bat movements, and doors and gates were often mentioned in descriptions of Wyandotte Cave. The first cave gate was erected in 1843 (Hov-

[1]*Deceased.*

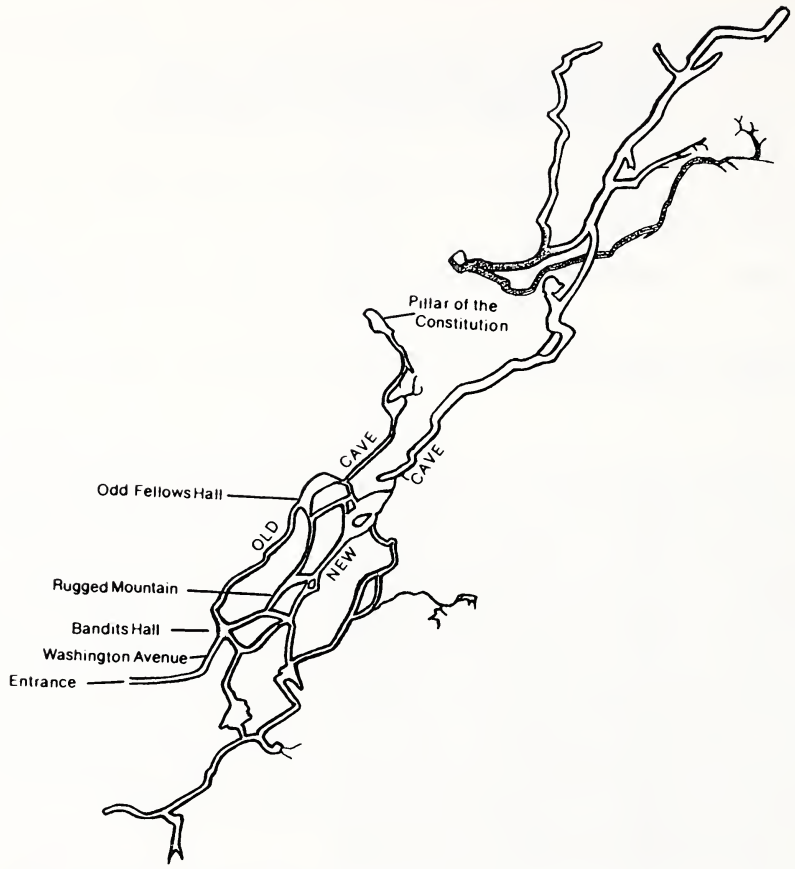

Figure 1.—Map of Wyandotte Cave (not to scale). The distance from end to end of the area shown is approximately 1.4 km.

ey 1880, 1882). It was just a fence in front of the cave entrance to keep out cattle that used the cave to cool off and to lick epsom salts from the rocks. Two doors were mentioned in early descriptions, an outer wooden door at the entrance, and a locked wooden door in the area of the entrance to the "New Cave" (Burnet 1851; Hender 1853). It was apparently removed about 1880 (George 1990). In 1858, a new door was built. Collett (1879) and Hovey (1882) said the outer door was now 6 m wide and nearly 2 m high. Apparently a half-meter of earth had been excavated from the floor and some of the talus removed. In 1970, the fourth gate was installed. It was a modern iron bar gate and was about 25 m into the cave. The last and present gate was constructed in 1991. This latter gate was designed to give bats good access into and out of the cave.

In November of 1850 Norman J. Coleman and party slid down a slope at the end of what is known as Bandits' Hall. They followed air flow and removed some rocks and gained entrance to a "New Cave." With the addition of the New Cave, the known size of the cave had more than doubled and now included very large rooms and prominent formations.

Walsh (1911) reported that when the original outer door was first installed, the cave housed a large colony of bats. There was only one known entrance, and when the door barred their exit "they flew blindly against it and were killed by thousands." Blatchley (1899) reported that "tens of thousands" of little brown bats, *Vespertilio subulatus*, were found in Odd Fellows' Hall. It is likely these bats were mostly, or all, Indiana myotis, although no concentration of bats has been found in this portion of the cave in recent years. This conclusion is supported by his description, which said they congregate very closely together. The total population of In-

diana myotis in Wyandotte Cave was estimated at 10,000 in 1953. That is the largest actual number of bats reported earlier (Mumford & Whitaker 1982), although there were claims that clusters of bats as large as a grand piano were once found in the cave. A grand piano at roughly 2.5 × 1.4 m would be about 3.5 sq. m, which would equate to up to 18,235 bats, using the estimate of Clawson (2002) that Indiana bats can occur at densities up to about 2510 per sq. m.

Although the cave is constantly visited, it presently contains numerous bats, mainly in winter, and is a major hibernaculum for the endangered Indiana myotis, *Myotis sodalis*. Further, the hibernating population of this species has grown; and it is now the third largest population of hibernating Indiana myotis in the state. Only Twin Domes and Ray's Caves have larger populations. We have a great deal of information about the bats that use Wyandotte Cave collected from 1953 to the present, mostly by mist-netting the entrance and by examining hibernating bats inside. The purpose of this paper is to summarize available information on the bats of Wyandotte Cave for use now and in the future.

METHODS

Cope and associates used two methods to study the bats of Wyandotte Cave. They entered the cave 28 times in the months of October through June between 1961–1983 to assess bat populations. They identified all the bats they observed (Table 1). To assess swarming, a mist-net was used to cover the entrance of the cave, and all bats were identified and their sex was determined. Data from 61 of these samples are presented here (Table 2). Mumford and Whitaker (1982) mist-netted the entrance to Wyandotte Cave all night on 23–24 August 1974 to assess the swarming population there. Whitaker, Todd Webb and Konnie Thessin did a similar all-night assessment 20 September 2001, although they used a 4 × 5 foot (1.2 × 1.5 m) harp trap rather than a mist-net. Also, second author (VB) netted the entrance in the spring of 1980 (22–23 May).

Brack and Dunlap (pers. comm.) entered Wyandotte Cave during every other winter from 1981 to 2001 to assess bats, particularly Indiana myotis, hibernating there (Table 3). This was part of the population assessment of the Indiana myotis by the Indiana and Gray Bat recovery team. However, only a small portion of the cave is surveyed for bats during their biannual survey (Fig. 1). Indiana DNR personnel (pers. comm.) indicate that there are rarely, or never, concentrations of bats in winter beyond the area of the cave surveyed.

RESULTS

Nine of the 12 species of bats of Indiana were seen during this work. The only species not included that occur in Indiana were Rafinesque's big-eared bat, *Corynorhinus rafinesquii*, the hoary bat, *Lasiurus cinereus*, and the evening bat, *Nycticeius humeralis*. The big-eared bat is considered accidental in Indiana, the hoary bat very seldom enters caves, and the evening bat has never been taken in caves. The nine species are discussed below.

Bats observed during 28 trips inside Wyandotte Cave between 1953–1981 (Table 1) included all nine species of bats taken there. However, only four species were taken regularly: *Myotis sodalis*, *M. lucifugus*, *Eptesicus fuscus* and *Pipistrellus subflavus*, in order of decreasing abundance. Only 1–3 individuals of the other five species were taken. Some of the trips (Table 1) were at times other than winter, the hibernating season. These trips were in October, April, and June; but, as would be expected, few bats were present at these times, and most were undoubtedly males.

Sixteen of these trips were taken in winter (December–March). *Myotis sodalis* was again the most abundant species, averaging over these times from 20–2141. *Myotis lucifugus* was second, ranging from 0–240. *Eptesicus fuscus* ranged from 0–45 and *Pipistrellus subflavus* from 0–19.

During 78 mist-nettings between 1961–1978 (Table 2), seven species of bats were rather regularly taken by mist-netting the entrance: *Myotis lucifugus*, *M. sodalis*, *Eptesicus fuscus*, *Pipistrellus subflavus*, *Myotis septentrionalis*, *Lasiurus borealis*, and *Myotis grisescens* (13 individuals of this latter species were taken).

The entrance to Wyandotte Cave was mist-netted on 22–23 August 1974 (all night) by Mumford & Whitaker (1982). They caught 399 bats of seven species, including 133 little brown myotis, 100 big brown bats, 91 Indiana myotis, 66 eastern pipistrelles, 6 northern my-

Table 1.—Bats observed on 28 trips on various dates inside Wyandotte Cave, 1953–1981, by Cope and associates. Means can be considered as estimates of the number of bats using the cave during different periods. In addition, some species were taken in much lower numbers as follows: one gray bat (*Myotis grisescens*) was recorded in February 1967/69, one northern myotis (*M. septentrionalis*) on 21 June 1966, two southeastern myotis (*M. austroriparius*) in February 1963/66, one red bat (*Lasiurus borealis*) in February 1970/74, and three silver-haired bats (*Lasionycteris noctivagans*), all in February, two in the 1963–66 period and one in the 1970–74 period.

Date	No. trips	*Myotis sodalis*	*Myotis lucifugus*	*Eptesicus fuscus*	*Pipistrellus subflavus*
Oct. 1963/1964	2	40	2	3	1
$\bar{x}$		20	1	1.5	0.5
Dec. 1961/1963	2	2428	450	15	5
$\bar{x}$		1214	225	7.5	2.5
Dec. 1967	1	638	0	0	0
Feb. 1963/66	2	2343	382	20	4
$\bar{x}$		1171.5	191	10	2
Feb. 1967–69	3	863	321	89	6
$\bar{x}$		287.7	160.5	44.5	2
Feb. 1970–74	5	457	22	128	13
$\bar{x}$		91.4	4.4	25.6	2.6
27 Feb. 1971	1	2141	4	17	2
Feb. 1976–79	2	702	11	19	3
$\bar{x}$		351	5.5	18.5	1.5
March 1953/1955	2	536	0	0	0
$\bar{x}$		268	0	0	0
March 1957/1959	2	942	233	0	0
$\bar{x}$		471	116.5	0	0
March 1961–65	3	2271	718	0	0
$\bar{x}$		757	239.3	0	0
25 March 1977	1	273	0	0	0
16 April 1966	1	28	31	34	3
21 June 1966	1	1	7	0	0

otis, 2 red bats and 1 gray myotis. However, on 20 September 2001, the picture was very different. Four species of bats, totaling 1245 individuals, were caught, including 843 Indiana myotis, 392 little brown myotis, 9 eastern pipistrelles, and 1 big brown bat. Thus there was an increase in the total number of bats but the number of species decreased from 7 to 4. The greatest increase was in Indiana myotis (from 91 to 843), and the little brown myotis increased (from 133 to 392). The community composition also changed. The Indiana myotis was the third most abundant species in 1974, forming 22.8% of the total catch, but was first in 2001 with 67.7%. The little brown myotis was the most abundant species in 1974, forming 33.3% of the sample, but dropped to second (392 of 1245, 31.5%) in 2001. In 1974, the sample included four species comprising 16% or more of the sample: the big brown bat (25%) and the eastern pipistrelle (16.5%), besides the Indiana and little brown myotis. In 2001 only the latter two species formed more than 16% of the sample.

The third author (VB) mist-netted the entrance to Wyandotte Cave on 22–23 May 1980. Unfortunately there was rain during much of the night. Mist-netting occurred from 2100–2300 h, and again from 0335–0500 h. All bats taken (37 individuals) were males, including 28 Indiana myotis, three northern myotis, three big brown bats, one little brown myotis and one eastern pipistrelle. It would appear that these bats were using Wyandotte Cave as bachelor quarters.

Regular bi-yearly counts of bats in hibernation by Brack et al. (1991) began in 1981 and have occurred every year since (Table 3). Six species were observed. *Myotis sodalis* is by far the most abundant species and has continually increased, from 2152 in 1981 to

Table 2.—Mean number of bats captured by mist-netting entrance to Wyandotte Cave 79 times 1961–1978, plus two all-night samples, one on 23 September 1974 and one on 20 September 2001.

Date	No. of samples	*Myotis sodalis*	*Myotis lucifugus*	*Myotis septentrionalis*	*Myotis grisescens*	*Eptesicus fuscus*	*Pipistrellus subflavus*	*Lasiurus borealis*
March 1977–78	3	14.3	11.3	3.3	0	21.7	0.3	0.3
April 1963–66	4	31.3	126.5	1.5	0.25	22.3	19.7	0
April 1977/1978	6	48	19	1.80	0	11.2	5	0
May 1962	2	6.5	29.5	0	0	32.5	8.5	0
May 1978	7	10	11.4	0	0.1	12.3	14.4	0
June 1978	1	137	39	2	1	12	22	0
July 1966	4	30.3	66.5	0.25	0	12.3	20.5	0.75
August 1961–1966	23	29.8	277	2	0.3	28.8	58.2	1.1
August 1974–1976	2	50.2	95.5	5.5	0.5	56	47	1
September 1961–1964	21	82.6	193.2	1.1	0.14	5.1	33	0.7
October 1963–1964	3	163	95.3	0.3	0	20	8.7	0
October 1968	1	12	0	0	0	0	0	0
All Night Sampling, 1974 and 2001								
22 August 1974	1	91	133	6	1	100	66	2
20 September 2001	1	843	392	0	0	1	9	0

28,584 in 2001. The other three regularly-occurring species are indicated in decreasing order as *Myotis lucifugus*, *Eptesicus fuscus*, and *Pipistrellus subflavus*, but all of these occur in relatively low numbers. The only other two species observed were *Lasionycteris noctivagans* (n = 2) and *Myotis septentrionalis* (1). The total number of bats during these observations increased each year and ranged from 10,344 to 28,584. These can be compared to the 16 earlier trips taken in winter (Table 1). The number of bats observed during these trips ranged from 91 to 2271, with the earlier counts being much lower.

SPECIES ACCOUNTS

Red bat, *Lasiurus borealis*.—The red bat is solitary and lives among the foliage. It is almost never found inside of caves, and none were found inside Wyandotte Cave (Table 1). However, 43 red bats were taken in mist-nets at the entrance to the cave. The fact that none were found inside the cave would seem to indicate that the bats merely swooped into the entrance and then flew back out. In an attempt to determine why red bats were netted at the cave entrance, we separated them by month and sex. If the majority were netted there in autumn (August–October, and perhaps in early

Table 3.—Bats counted during biyearly counts in hibernation in Wyandotte Cave by Brack and Dunlap.

Year	*Myotis sodalis*	*Myotis lucifugus*	*Eptesicus fuscus*	*Pipistrellus subflavus*	Other species
1981	2,152	6	11	2	0
1983	4,550	0	0	0	0
1985	4,627	21	2	1	0
1987	6,681	275	12	2	0
1989	10,344	8	32	14	*L. noctivagans*—1
1991	12,994	15	11	21	*L. noctivagans*—2
1993	17,304	12	38	4	0
1995	23,878	30	53	18	0
1997	25,424	70	24	32	0
1999	26,854	268	17	13	0
2001	28,584	272	10	16	*M. septentrionalis*—1
Total	163,392	977	210	123	
$\bar{x}$	14,853	89	19	11	

Table 4.—Locations of Indiana bats *Myotis sodalis* in hibernation in Wyandotte Cave during the past seven regular biyearly counts by Brack and Dunlap.

	Entrance		Washington Avenue		Rugged Mountain		Rest of survey area		
Year	No.	%	No.	%	No.	%	No.	%	Total
1989	23	0.2	8,894	83.1	781	7.6	946	9.1	10,344
1991	12	0.1	11,301	87.0	1,658	12.8	23	0.2	12,994
1993	12	0.1	11,995	69.3	3,719	21.5	1,578	9.1	17,304
1995	84	0.4	20,644	86.5	1,897	7.9	1,253	5.2	23,878
1997	6	0.0	21,994	86.5	3,365	13.2	59	0.2	25,424
1999	1,896	7.1	21,520	80.1	3,222	12.0	216	0.8	26,854
2001	1,531	5.4	19,662	68.8	7,167	25.1	224	0.8	28,584

spring) it could mean they came in search of mates. One male was there in March, but then no red bats were netted until the end of July. Most red bats at the cave entrance were taken in August or September (25 ♂ and 15♀). Since this is the time of dispersal and mating, perhaps a few do use the cave entrance as a place for finding mates.

Silver-haired bat, *Lasionycteris noctivagans*.—A few silver-haired bats hibernate in caves in Indiana, but otherwise silver-haired bats are found in Indiana only during spring and fall migrations (March through early June; and late September through November). Six were seen during this work. One male was found in hibernation inside Wyandotte Cave on 24 February 1973, and three were seen during the winter counts, one on 9 February 1989 and two on 1 February 1991, at locations near the entrance. The other two, a female (26 March 1977) and a male (31 March 1978) were taken by mist-net.

Southeastern myotis, *Myotis austroriparius*.—Only one southeastern myotis was found during this work. It was hibernating in Wyandotte Cave on 12 February 1966. Southeastern myotis were regularly found in caves in Lawrence and Greene Counties (especially Donnehue's Cave) through the late 1970's with the last one being found on 4 February 1977. However, there have been no verified records since. This species appears to have become extirpated in the state.

Gray myotis, *Myotis grisescens*.—The only maternity roost(s) of gray myotis known in Indiana is in Clark County where the bats spend the summer in a quarry at Sellersburg. Gray bats were first discovered in Clark County by Cope (unpubl. data) by netting in Muddy Fork Creek just north of Sellersburg in 1980; but the actual roost in the quarry was not discovered until 1982 (Brack et al. 1984), when about 400 gray bats were present. This population near Sellersburg has increased to nearly 4000 bats. Pruitt et al. (1997, 1998) discovered gray myotis at the Charlestown army installation, about 5 km NE from Sellersburg, but the specific site of the Charlestown roost has not been found. These roosts are assumed to be part of a single colony since bats have been documented moving between the two localities.

Only one gray bat was taken inside Wyandotte Cave by Cope and associates (Table 1). It was found on 10 February 1967. However, 12 gray bats (including 4♂ and 4♀) were mist-netted near the cave in April (1), May (1), June (1), August (6), and September (3). Autumn is the time of dispersal, which probably explains the larger numbers in August and September. These bats could have come from Sellersburg; but it is more likely that they were from colonies from northern Kentucky, which are much closer than those at Sellersburg. In addition, four male gray myotis were found in Wyandotte Cave on 26 July 1989. They were among a sample of 23 bats (the other 19 were male Indiana myotis) from two clusters, totaling an estimated 600 bats near a formation called The Pillar of Constitution (Fig. 1). Another two male gray myotis were captured in April 1991 at Wyandotte Cave during spring emergence. The occurrence of gray bats at Wyandotte Cave is not surprising since Whitaker & Gummer (2001) netted gray myotis at seven sites along the nearby Ohio River between Spencer and Clark Counties.

Eastern pipistrelle, *Pipistrellus subflavus*.—This species hibernates in caves and mines usually not more than 180 km from the summer roost. It enters the hibernaculum early in the autumn and exits in April and May. Forty-four pipistrelles (1.1 per trip) were recorded inside the cave by Cope and associates (Table 1). However, pipistrelles are solitary, and are often found deeper in the cave than are other species. Netting pipistrelles yielded 307 individuals in 19 counts in April (16.1 per netting). Males outnumbered females among exiting pipistrelles. This might indicate that males remained near the entrance longer, and perhaps summered there, whereas females left to establish maternity colonies. Seventy individuals were netted at the cave entrance in June and July, but all were males, probably indicating that this was a bachelor roost for a small number of pipistrelles. Numerous nettings in August and September yielded numerous pipistrelles (mean = 59.6, range of 0–171 in August; mean = 33.3, 1–79, 21 nettings in September). There were many more males than females at this time. These pipistrelles were probably migrating through or returning to the cave for hibernation. In the case of the latter, the females were apparently quickly entering the cave for hibernation. The males were spending more time at the cave, presumably for mating purposes; thus their numbers were higher than those of the females (males: mean of 8.7 per night of mist-netting, females 1.3 per night of mist-netting).

Northern myotis, *Myotis septentrionalis*.—*Myotis septentrionalis* hibernates in caves and is common in Indiana. However, it is seldom found while hibernating. Only one bat of this species was found in Wyandotte Cave by Cope and associates (Table 1) during 30 visits in winter, and only one was recorded by Brack and Dunlap between 1981–2001. The fact that few individuals were seen hibernating in Wyandotte Cave does not mean that this species does not hibernate there. The fact that only 142 individuals were captured during 77 nettings (n = 1.8 bats per netting) does indicate that few northern myotis use Wyandotte Cave either for swarming or for hibernation. The largest number seen in any one trip was 10 August 1965 when 10 males were captured. Only six were captured in an all-night netting 22 August 1973, and none were captured in an all-night sampling on 20 September 2001. Whitaker & Rissler (1993) found numerous bats swarming and much winter activity at Copperhead Cave, a mine in Vermillion County, Indiana; but none were seen in hibernation there. However, Whitaker & Rissler (1992) estimated that about 900 northern myotis hibernate in tiny cracks in this mine.

Big brown bat, *Eptesicus fuscus*.—The big brown bat is the most abundant bat in Indiana, in great part because it has taken advantage of man-made structures other than mines. Most maternity colonies are in buildings or other structures (we know of only two maternity colonies in trees). One maternity colony is in a building in the town of Prairie Creek, Vigo County, Indiana; and before parturition, adults fly 5 km to a 650 ha wooded bottomland area near the confluence of Prairie Creek and the Wabash River. After the young are volant, the bats form a "post-maternity colony" under loose bark of trees at the Prairie Creek site. A few big brown bats hibernate in caves and mines in winter, usually near the entrance in small numbers. We estimate, probably generously, that not more than 15,000 of the state's 1,200,000 big brown bats hibernate in caves and mines. The rest hibernate in buildings. We recorded only 326 big brown bats (Table 1) hibernating in Wyandotte Cave in the 30 visits (mean = 12.2 bats per visit), although there were undoubtedly more, since Wyandotte Cave is of large size. In caves and mines the big brown bat hibernating population is usually male biased. However, this sample consisted of 161 male and 165 females. At Wyandotte Cave relatively few big brown bats were swarming, totalling only 179 bats in the 77 visits (mean = 2.3 per visit). Likewise, Brack & Dunlap (1999) recorded few big brown bats (0–38) in Wyandotte Cave in the 1981–2001 period.

Indiana myotis, *Myotis sodalis*.—The most abundant hibernating bats in Wyandotte Cave are the Indiana and little brown myotis. However, the numbers of Indiana myotis are consistently larger than those of little brown myotis as indicated by all winter dates (Tables 1, 2). A total of 3955 Indiana myotis was observed at Wyandotte Cave during 77 mist-netting sessions between 1961 and 1978 (Table 1), mean = 51.4 per session (range 0–449). The numbers inside the cave in December through February 1960–1975 ranged from 0–2164 (mean = 615.3).

The population of hibernating Indiana myotis in Wyandotte Cave has increased greatly over the past several years. There were 1372 bats counted in February of 1979, and the numbers have increased every year since the biannual surveys began—from 2152 in 1981 to 28,584 in 2001 (Table 3). Thus, populations remained relatively low until about 1980, when they began to increase dramatically, until Wyandotte Cave currently has the third largest wintering population in the state after Twin Domes and Ray's Caves, and followed by Jughole and Batwing. The largest known population of this species was Pilot Knob Mine, an iron mine in Missouri, which is now too dangerous to enter. It was estimated to hold 139,000 Indiana myotis in 1975–78, but Clawson (pers. commun.) estimates that it has declined by as much as 75% since.

Apparently the reason hibernating populations in Wyandotte Cave of *Myotis sodalis* have changed so over the past 40 years, and have increased over the past 20 years, is because of structural changes at the cave (Richter et al. 1993). A gate was built in 1977 to keep people out at unauthorized times. However, the gate increased cave temperatures, which was detrimental to the bats. A new gate was constructed in July 1991 (Johnson et al., in press). It improved air flow, helped cool the cave, and allowed bats in. The number of bats hibernating in Wyandotte Cave immediately began to increase. It is clear that the early gate was detrimental to Indiana bats and that the new gate favored them.

The Indiana myotis normally hibernates at lower environmental temperatures (2–6° C) than other species, and also does best when temperatures are stable. Wyandotte Cave is very large with stable temperatures of about 10° C. This temperature is more appropriate for other species, yet many Indiana myotis hibernate there. For example, in 1991 the temperature in the entrance area was 5° C, but only 1896 Indiana myotis hibernated there. Most of the bats were in the Washington Avenue area (21,520 bats, temperature 8–9° C) and Rugged Mountain area (3222 bats at 10° C). Fewer bats (216) were counted in area D (the rest of the survey area) where the temperature was 12° C. The temperature further inside the cave from where the Indiana myotis hibernated was 13° C.

The number and percentage of Indiana myotis using various portions of the cave have varied over time (Table 4). The Washington Avenue area has always contained the greatest number and percentage, varying from 68.8% (2001) to 87% (1991). The Rugged Mountain area has contained from 7.6% (1989) to 25.1% (2001) of the hibernating population. The entrance usually has had low numbers and less than 1% of the population, but in 1999 and 2001 it contained 1896 and 1531 Indiana myotis (7.1% and 5.4%, respectively, of the population).

Little brown myotis, *Myotis lucifugus*.— The little brown myotis is the second most abundant bat hibernating in Wyandotte Cave, yet its numbers usually are not large. It is most abundant during hibernation in Area D. The mean number hibernating in Wyandotte Cave ranged from 0–225 between 1953–1980. It is noteworthy that, although the numbers of Indiana myotis hibernating in Wyandotte Cave were larger than the numbers of little brown bats from 1953–1980, the number of little brown myotis mist-netted during that period was much larger than that of Indiana myotis. The mean number of little brown myotis in the 77 nettings was 157.5, whereas it was only 51.4 for the Indiana myotis. However, in the last two decades (1980–2001), during the increase in the numbers of the Indiana myotis, numbers of little brown myotis hibernating in Wyandotte Cave were still lower, averaging 88.8 (range 0–275) per visit during biannual counts. It should be mentioned that during the winter hibernating period, individuals and small clusters of little brown myotis are more likely to use portions of the cave beyond those areas surveyed, and therefore may not be found and included in the counts. No mist-netting data are available for the period 1982 through 2000, but on 20 September 2001, the entrance to Wyandotte Cave was mist-netted, and at that time Indiana myotis outnumbered little brown myotis 843 to 392. The only other bats taken that night were nine eastern pipistrelles and one big brown bat.

DISCUSSION

It is not clear why Indiana myotis continue to do so well at Wyandotte Cave when the temperature in the areas where the bats hibernate is about 10° C, whereas their preferred hibernating temperature is about 2–6° (Hall 1962). We have attempted to help protect In-

diana bats by preventing winter visitations to hibernacula so that the bats would not be awakened and use energy that is supposed to see them through the winter. However, the bats themselves (at least *M. lucifugus*, and presumably other species) negate some of our efforts. They arouse periodically from hibernation, about once every two weeks for a few hours. During these arousals, which collectively total about 53 h, the bats expend about 75% of their stored energy (Thomas et al. 1990). During these arousals, the bats often move about, both as individuals and perhaps even in groups.

Disturbance is also an important factor; therefore, it has been general policy that Indiana myotis hibernacula are not visited during the hibernating season except once every other year to assess the bat population. However, Wyandotte Cave has been a major exception to the general policy. Through the winter of 2001–2002 there were guided tours through the cave, and these tours went directly under one of the main areas (Washington Avenue) where the bats hibernate. Thus, data from this cave suggest that the non-visitation policy is not necessary to conserve energy for successful hibernation, as the bats have continued to increase even in the face of regularly occurring visits, often several per day. However, non-visitation does prevent vandalism on hibernating bats.

Brack et al. (1991) reviewed alterations of cave morphology and airflow that have occurred in Wyandotte Cave. Richter et al. (1993) reviewed historical changes in the number of bats hibernating in the cave and related them to man-made changes that affected airflow. More bats apparently hibernate in the cave because of gating changes that improved airflow and are more bat-friendly. The 2001 survey was the fifth since a bat-friendly, angle iron gate was installed in 1991.

Albino bats are rare. However, during recent surveys, one albino Indiana myotis was observed in Ray's Cave during three consecutive biannual surveys (1985, 1987, 1989) (Brack & Johnson 1990). During the 1995 and 1997 surveys, a single white bat was found in Wyandotte Cave. No white bats were observed during more recent surveys.

The numbers and locations of big brown bats, little brown bats, and eastern pipistrelles have varied over the years. Big brown bats are typically found only near the entrance, and most little brown bats are usually found beyond the Rugged Mountain area.

It appears that the early gate installed at Wyandotte Cave was detrimental to the bats but that the new gate is satisfactory. Since the new gate was installed, the number of hibernating Indiana myotis has continued to increase. However, the number of little brown myotis hibernating there has decreased since that time. The reasons for the decrease in little brown myotis are unknown, but a similar decline was noted at Ray's Cave as the number of Indiana myotis increased.

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ACHIEVING RELEVANCE IN AN INTRODUCTORY ENVIRONMENTAL SCIENCE COURSE THROUGH PRE-SERVICE TEACHER ARTIFACTS

Amy L. Sheaffer: Department of Natural Resources and Environmental Management, West Quad 114, Ball State University, Muncie, Indiana 47306 USA

ABSTRACT. Beginning in the Fall 2002 semester, science departments at Ball State University were required to document content learned by pre-service teachers enrolled in science courses. This documentation was to be achieved by creating new assignments for science courses that matched state standards for teacher licensure. The purpose of this article is to discuss how one department initiated new assignments in an introductory environmental science course that not only served the needs of pre-service teachers but also were relevant to the science majors and non-science majors enrolled in the course. Three examples of assignments are discussed as they relate to finding common ground among students from different majors in an introductory course and for promoting scientific and environmental literacy.

Keywords: Pre-service teachers, environmental literacy, science education

In the Fall of 2002, Ball State University began a campus-wide initiative to document the content learned by pre-service teachers in science courses. Science departments across the university were mandated to create artifacts for pre-service teachers. Artifacts were defined as specific class assignments with corresponding grading rubrics that students would keep for future teaching portfolios. In the Department of Natural Resources and Environmental Management (NREM), an introductory environmental science course is taught that is required of the pre-service teachers in the life-science licensure program. These students are seeking a license in life science for teaching senior high school, junior high school, or both. They are required to take only one course from the department, namely NREM 101, "Environment and Society."

Two other sub-populations served by this course are science majors, e.g., in NREM or other programs, and non-science majors fulfilling a requirement in the university core curriculum. It is a challenging course to teach because of the variety of students it attracts and the need to make content relevant to students from different disciplines. This pattern of teaching science majors and non-science majors as well as pre-service teachers in the same course is not unique to NREM 101. Other science courses at Ball State University are also taught in this way, for example, Geography 101, Geology 101, Health Science 160 and others. Therefore, the overall goal in creating new assignments for the introductory environmental science course was to provide content appropriate to all students that also matched state standards for life-science licensure among the pre-service teachers.

The need for documenting science content learned among pre-service teachers had been clearly identified, and the start date had been established by university mandate. In preparation, I was assigned as the representative from the NREM Department to serve on the life-science licensure committee for the university and was responsible for developing the new artifact-assignments for the "Environment and Society" course. My intent was to create assignments that could be used by all faculty members teaching the introductory course. After the assignments were created, they were given to the other four instructors to obtain their feedback. As a group, the faculty members responsible for the course had discussions about how the pre-service teacher artifacts required could ideally benefit all students in the class. Three new assignments were created based on two current debates in the environmental field that all instructors felt were important to cover, namely human population growth and global climate change. These particular topics also match well with pre-service teaching standards of the Indiana

Professional Standards Board (IPSB). The assignments were designed to be either laboratory or take-home assignments for use in the class.

SUB-POPULATIONS IN THE CLASSROOM

The three sub-populations in the introductory environmental science course are now considered and discussed based on a review of relevant literature.

Pre-service teachers.—A literature review shows that other faculty teaching introductory science courses have evaluated course content and considered the needs of pre-service teachers. Starr (1995) explored cooperative learning in a geology course for pre-service elementary-school teachers. Part of her motivation was to find a user-friendly method that encouraged women to enroll and remain in the sciences. She tested cooperative learning to see what results it would have on achievement in the course. Results suggested that interactive, group learning techniques helped students grasp geologic concepts such as minerals and rock formation, plate tectonics, and geologic time. Her students were better able to learn course content when they interacted with each other. The content in Starr's geology course applies to the introductory course "Environment and Society" being discussed in this article, because similar topics are covered.

Heppert et al. (2002) discussed techniques for enhancing introductory chemistry courses for pre-service teachers at the University of Kansas. They suggested problem-based, inquiry learning units and interdisciplinary elements to increase the relevance of the course content for teacher education students.

The introductory environmental science course, NREM 101, is taught by several faculty members in the department and serves a large number of students each semester. Even though only a few students in the class are teacher education majors, faculty have the important task of developing the environmental literacy of these science teachers-in-training. An example of the language that was included in the syllabus for addressing pre-service teachers is as follows:

> *New for Fall 2002:* Teacher education majors are advised to document what they have learned in their classes to show that content acquired matches state standards for the teaching license being sought. If you are a teaching major, or if you think you will ever decide to be teaching major, you should save each assignment (called an artifact) given by your instructor for a teaching portfolio.

The instructors of this course invest in the development of pre-service teachers who will teach many junior high and high school students in the future. Ambers (2002) offered this perspective:

> College represents the last opportunity the educational system has to impress students with the importance of learning science, and it is the training ground of future science teachers.

For these reasons, pre-service teachers are an important sub-population in an introductory science course and their need for meeting state education standards relates to decisions about course content by faculty members.

Science majors.—Science majors take the introductory environmental science class as a foundation course for their science programs. They need to establish an understanding of basic scientific principles on which they can build as they continue in their majors and areas of study. Science majors must gain competencies with scientific concepts as they relate to environmental issues, such as the states of matter, the basic process of photosynthesis, population models, etc. Hoots (1999) argued for inquiry-based science for science majors as well as the need for adding a humanistic side to science. By adding a humanistic side to the classroom, science majors can appreciate the contributions of other fields, e.g., creative writing and communication for teaching others, especially the general public, about science.

Examples from the literature discussed thus far have suggested that both pre-service teachers and science majors can benefit from interdisciplinary perspectives applied to environmental issues. They need to understand how non-science disciplines affect environmental decision-making. This is the type of perspective that students who are non-science majors can bring to an introductory science course.

Non-science majors.—The needs of non-

science majors are particularly deserving of some consideration because, for many, this class serves as one of a relatively small number of science courses they take at the university. Further, non-science majors are the largest sub-population in the introductory environmental science course being discussed. In the Fall 2002 semester, 90% of students enrolled in the "Environment and Society" course were non-science majors. For these students, the primary goals of this course are to promote their understanding of basic science to improve their scientific and environmental literacy and to help them see the relevance of understanding environmental issues in their daily lives. One means of achieving these goals is to show the bridges between science and other fields, such as the arts and humanities. This was the intent of an interdisciplinary course at Northern Arizona University linking art, math, and chemistry, designed specifically for non-science majors (Kelley et al. 2001). As an example of such a link, students considered how scientific methods could relate to proving artwork as authentic or forgery, for example, using chemical analyses. As another example, the Workshop Biology course at the University of Oregon was designed to show the philosophy of *science in context*, meaning . . . the role science plays in society has personal and social implications. These examples suggest that an interdisciplinary approach may prove beneficial for non-science majors.

There are common goals for all students in an introductory environmental science course. Non-science majors as well as science majors and pre-service teachers should all be better able to decipher the current debates that surround environmental issues after they leave an introductory course. They should improve their critical thinking skills as they learn the content. For example, they should be more knowledgeable when reading a newspaper about an environmental issue, such as global climate change or human population growth, and should be able to detect biases on both sides of a particular debate. Regardless of their majors, students in the course should be able to apply basic science to environmental issues. The pre-service teacher artifacts were designed to accomplish these common goals for all three sub-populations in the classroom.

ESTABLISHING A SENSE OF RELEVANCE

Environmental issues can be taught in such a way that the science behind them is relevant to the general public; for example, environmental quality affects our personal health, the quality of our food and water supplies, and energy for our homes and cars (Udovic et al. 2002). Moseley (2000) argued that environmental literacy requires a progression, or continuum, of learning: nominal literacy is the ability to recognize basic environmental terms; functional environmental literacy is the ability to bring ideas together in forming action positions on environmental issues; and operational environmental literacy allows students to choose a course of action from among alternatives. Students should also develop "the skills needed to make decisions where science, technology, and human values interface" (Berkson & Harrison 2002). This is a progression that needs to develop over time. Students in the first few weeks of an introductory environmental science course are at the early stages of Moseley's continuum, i.e., learning basic environmental concepts. As the semester progresses, they should begin to bring concepts together and have a better understanding of the complexity of environmental issues. Near the end of the course they begin to think about action alternatives in their everyday lives; however, this type of higher level thinking has to come with time throughout their college careers and on into adulthood. Thus the stage of operational environmental literacy, or the end of Moseley's continuum has to develop beyond an introductory course into their adulthood. The early stages of environmental literacy can begin in an introductory course.

Wallace (2002) argued that environmental science should begin as early as possible in a student's college program. He suggested that environmental literacy should be nurtured in the freshman year. In this way, students can have a foundation in environmental concepts with which to balance the conventional wisdom of their disciplines and majors that they pursue in subsequent years. He also implied that students should be taught to consider many different points of view surrounding environmental issues.

Salmon (2000) echoed this suggestion. He

argued that environmental issues, by their vary nature, should be open to debate, and that this debate is the kind of material ripe for student contemplation. Environmental education curricula should present environmental issues from many perspectives, i.e., material should be based less on trying to mold behavior solely from one perspective, and more on trying to build knowledge, awareness and critical thinking. He noted the following:

> Giving students oversimplified answers to complex problems does not make them effective citizens and can even breed cynicism . . . Even among environmentalists, there is no consensus on what constitutes 'sound' environmental choices . . . on what are the most effective solutions for many environmental problems, and to state or imply there is constitutes a form of miseducation.

In addition, learning science and the process of scientific inquiry involves more than just memorization of facts. Other perspectives are that science is a way of knowing, i.e., "science as inquiry," or that science has context in our personal and social lives (Udovic et al. 2002). Hoots (1999) suggested that the humanistic side of science should be incorporated into introductory courses. As she aptly described:

> Too often we tend to confuse information with understanding. Knowledge without meaning or understanding is nothing more than empty rhetoric. To accommodate the information, it must be meaningfully woven into stories that can be related to our experiences and that will enable us to expand our perceptions.

The point to be made here is that effective teaching of environmental science incorporates a degree of relevance, i.e., a connection between course content and a student's understanding. Ham (1992) argued that environmental information is relevant when it is meaningful and personal. Being meaningful implies that it has context because the information connects to previously acquired knowledge, while being personal suggests that the information is something students feel is important. Berkson and Harrison (2002) suggested: "achieving scientific literacy requires students to understand how science is applied in the real world." Thus, critical thinking and a sense of relevance was deemed important for all three sub-populations in the environmental science course and guided the introduction of the new artifact-assignments.

Artifacts as assignments for all students.—In creating pre-service teacher artifacts that would serve as assignments for all students, I chose two broad topics that are currently debated in the environmental field. I was responsible for developing the artifacts for the department as a whole and then I asked for feedback from other instructors in the course. Other instructors made suggestions for modifications to the assignments. The following artifacts were then introduced as assignments in several sections of the "Environment and Society" course in the Fall 2002 semester: 1) Human Population Growth, 2) the Carbon Cycle, and 3) Global Climate Change. Together, these assignments were worth between 10% and 15% of the overall grade for the class.

Human population exercise.—Students performed this assignment in a computer laboratory. The instructor began the class with a discussion in which students proposed possible factors that could relate to population growth. The instructor wrote students' ideas on the blackboard and discussed the basic debate over population growth. Students were then directed to the data files on their computers. They were allowed to work in pairs but had to each hand in their own worksheets at the end of the lab session. They were given data from the Population Reference Bureau (http://www.prb.org/) and the World Resources Institute (http://www.wri.org/) describing factors that relate to human population growth and natural resource supply organized in an MS Excel spreadsheet. For each variable that they graphed or analyzed they compared other countries to the United States. This was one strategy for achieving relevance for the students, i.e., by having them think about issues inside and outside their own country. Students compared 25 countries to the United States in terms of the following factors: the rate of natural increase, doubling time for the population, population density, fertility rate, hectares of forest land, and hectares of cropland per 1000 people. Students were required to do basic data analysis and create graphs to illustrate their results. Finally, they were asked to label

Table 1.—Population Growth—Standards of the Indiana Professional Standards Board addressed in the Human Population Exercise. (http://www.state.in.us/psb/standards/ScienceContStds.html)

Population Growth
■ Populations grow or decline through the combined effects of births and deaths, and through emigration and immigration. Populations can increase through linear or exponential growth, with effects on resource use and environmental pollution.
■ Various factors influence birth rates and fertility rates, such as average levels of affluence and education, importance of children in the labor force, education and employment of women, infant mortality rates, costs of raising children, availability and reliability of birth control methods, and religious beliefs and cultural norms that influence personal decisions about family size.
■ Populations can reach limits to growth. Carrying capacity is the maximum number of individuals that can be supported in a given environment. The limitation is not the availability of space, but the number of people in relation to resources and the capacity of earth systems to support human beings. Changes in technology can cause significant changes, either positive or negative, in carrying capacity.

a world map to indicate regions with higher rates of population growth and those with lower growth rates.

Pre-service teachers: For pre-service teachers, this artifact-assignment meets two specific IPSB standards for the teacher licensure in the life sciences: 1) Population Growth, and 2) Natural Resources. Pre-service teachers were instructed to keep this assignment for their future electronic portfolios. The state standards addressed by this activity are shown in detail in Tables 1 and 2.

Relevance for all students: The Human Population Growth assignment was used to promote the environmental literacy among all students. The assignment served to promote greater awareness of geographic regions of the world as they relate to the concepts of developing and developed countries. Students learned how population growth patterns and access to natural resources help to define nations into one of the two categories. Students became familiar with viewing spreadsheets of data and creating graphs and charts to illustrate basic population concepts, e.g., population density. Students were required to use their critical thinking skills to try to detect patterns in the data. For example, they were asked to summarize the assignment with the following: Based on your answers, what in general do you observe? In which areas of the world is the population growing most rapidly? In which areas of the world is it growing more slowly? Do different areas of the world vary by their access to resources, like forests and cropland? Are there any patterns between population growth and access to resources? Write a two-paragraph summary explaining the trends you have observed in the data provided for this assignment.

Carbon cycle and global climate change exercises.—Another important debate covered in the environmental science course is that of global climate change. However, before students can understand the complexity of the debate, they first need to understand how carbon cycles through the Earth's systems. Thus two assignments were created—a take-home assignment on the carbon cycle and a laboratory simulation assignment on global climate change. They addressed the Natural Resources standards for pre-service teachers (Table 2) as well as other IPSB standards for Geochemical Cycles and Environmental Quality.

In the Carbon Cycle activity, students addressed several scientific concepts in a take-home assignment. They discussed the states of matter and the reservoirs for carbon compounds on Earth, namely living resources (organisms), soil and mineral resources (solid Earth), air resources (atmosphere), and water resources (freshwater and saltwater reservoirs). This activity encouraged them to think of carbon compounds as part of their everyday lives, e.g., carbon dioxide that they exhale, carbohydrates as a source of food, hydrocarbons which provide fuel for their cars, for electricity and heat in their homes, and limestone (calcium carbonate) as a raw material for buildings. In their assignments, students applied basic science principles to the cycling of carbon through photosynthesis and aerobic respiration, the formation of limestone and fossil fuels, and the production of carbon dioxide from fossil fuel use, which is a concern in the global climate change debate.

Table 2.—Natural Resources—Standards of the Indiana Professional Standards Board addressed in the Human Population Exercise. (http://www.state.in.us/psb/standards/ScienceContStds.html)

Natural Resources
■ Human populations use resources in the environment in order to maintain and improve their existence. Natural resources have been and will continue to be used to maintain human populations.
■ The earth does not have infinite resources; increasing human consumption places severe stress on the natural processes that renew some resources, and it depletes those resources that cannot be renewed.
■ Humans use many natural systems as resources. Natural systems have the capacity to reuse waste, but that capacity is limited. Natural systems can change to an extent that exceeds the limits of organisms to adapt naturally or humans to adapt technologically.

A computer simulation model was given as an in-class laboratory assignment following the completion of the students' carbon cycle assignments. Hothouse Plant is a global climate change simulation available through EME Corporation (2003). For this assignment, students worked at computer stations in the departmental computer classroom. They explored greenhouse gases and did experiments in which they changed the concentrations of greenhouse gases through data manipulation. They predicted changes in sea level and temperature and related these numbers to specific cities and regions of the United States, for example, dry land loss in coastal regions. They also explored the predicted effects of volcanic aerosols and solar radiation on changes in atmospheric temperatures. The simulation model, combined with the carbon cycle assignment, was designed to help students reflect on the impacts of daily human activities as they relate to the debate over global climate change.

Assessment.—Grades were compared for science and non-science majors in one section to assess the new assignments introduced in the Fall 2002 semester. For the Human Population Growth assignment, the average grade for non-science majors (n = 23) was 86%, while the average grade for the science majors and pre-service science teachers combined (n = 6) was 92%. For the Spring 2003 semester, the average grade for non-science majors (n = 23) was 85%, while for the science majors (n = 5) it was 90%. The non-science majors may be at a slight disadvantage in this assignment. Students gave the instructor feedback on the assignment. Verbal comments suggested that making graphs of data was unfamiliar to both science majors and non-science majors, therefore it was important for all students to practice these skills. Students indicated that they thought this was a challenging assignment.

When examining the Carbon Cycle assignment grades for Fall 2002, science majors and non-science majors scored on average an 88%, indicating that the content is suitable for both groups. For the Global Climate Change laboratory assignment, the average score for non-science majors was 90% versus 89% for the science majors. In general the comparison of grades suggests that the basic science content for the three artifact-assignments is suitable for students in the class regardless of their majors.

Faculty members teaching the course also assessed the assignments for suitability in their individual courses. For those with sections larger that 32 students, the Human Population Growth and Global Climate Change exercises designed for the computer laboratory can create a problem because there are only 32 computer stations in the department. Instructors in these larger sections have discussed using the assignments as take-home assignments, giving the responsibility to the students of finding computer facilities on their own. One problem with this approach is that students sometimes like to ask questions of the instructor while completing the assignment, such as in a laboratory setting. These and other issues related to the new assignments are continuing to be addressed in discussions among instructors of the course about using them in the future.

DISCUSSION

A university-wide pre-service teacher initiative began in the Fall of 2002. There was a need for documentation of science content learned by pre-service teachers pursuing a life-science licensure. Artifact-assignments connected to state standards were the requirements for achieving this goal. However, arti-

facts were also designed to serve an additional purpose, namely as assignments for all students enrolled in the introductory course. When the new assignments were proposed to the five instructors of the course, an important issue was the freedom for each instructor to follow a preferred procedure. In assessing the implementation of three new assignments in the "Environment and Society" course, the five instructors who teach the class decided to use them in very different ways. Some instructors used all three assignments, while other instructors used one or two of the assignments. Further, some instructors required the new assignments of all students enrolled in the class, while others gave the artifact-assignments only to the pre-service teachers as extra assignments to be completed outside of class. Thus, there was healthy debate surrounding how these new assignments should be used in the department.

The strategy that has been discussed throughout this article has been to use the pre-service teacher initiative at Ball State University to introduce assignments for all students in an introductory environmental science course to develop their environmental literacy. The content of the assignments was intended to incorporate basic scientific principles that U.S. citizens should know to understand environmental debates and that was deemed appropriate regardless of the students' majors. The discussion presented above was one possible strategy for combining the goals of a science department for the students it serves with the goals of a university for pre-service teacher development.

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MINUTES OF THE MEETINGS OF THE INDIANA ACADEMY OF SCIENCE 2002

COMBINED MEETING OF THE
EXECUTIVE COMMITTEE
AND THE COUNCIL
Butler University
12 April 2002

President Terry West called the meeting to order at 2:00 PM with 21 members in attendance. Those attending were: Dick Conklin, Jeff Hughes, Dan Webster, Damian Schmelz, Uwe J. Hansen, Frank Guthrie, David Daniell, Donald Ruch, Edwin Squires, Jim Curry, Jim Berry, Holly Oster, Marcia Moore, William Gommel, Ruth Howes, Edward Frazier, Terry West, Robert Waltz, Marcia Gillette, Duvall Jones, Robert Dale.

Minutes from the Fall 2001 meeting were approved as proposed. Motion by Uwe Hansen, seconded by Ed Squiers. Secretary Nils Johansen was absent so Nelson Shaffer took notes.

Treasurer Ed Frazier presented an account summary of the budget with income from dues, interest, and contributions of $12,909.60 and additional income from publications and returned grant money and a grant from the American Association for the Advancement of Science for a total of $32,222.09. Operating expenses totaled $66,668.22, leaving a deficit of $34,446.13. An IRS audit of Academy book found that all was in order.

Executive Director Nelson Shaffer reported paid membership to date of 706 and gave a breakdown of membership categories. He also outlined initiatives to increase membership and public visibility of the IAS.

Jim Berry, Editor of the *Proceedings*, reported that Volume 110 was published and that Volumes 108/109 should be available soon. They will contain papers about the Grand Calumet River region. The matter of page charges was discussed, but no motions were made.

Newsletter editor Uwe Hansen reported that newsletters were produced on time and within budget. He complimented the printer, mail service, and Marilyn DeWees for their efforts. Uwe called for contributions to the newsletter, especially research results. Discussion of formats, inserts, and putting the newsletter on the IAS website ensued.

President-Elect Robert Waltz deferred his comments until new business.

President Terry West called for abstracts for the Fall Meeting. He noted the early abstract deadline (August 26) and that meeting information is on the IAS website. He previewed the field trip to the reclaimed Kentucky Avenue landfill and the Martin Marietta Underground Mine.

Committee Reports

Frank Guthrie of the Foundation Committee reported on the three accounts and that total assets had decreased about 7½% since December 2000. The Foundation account stood at $6,751,000 on March 23. The Committee is working with account managers Merrill-Lynch to develop an investment policy with more diversity. The Committee wants to have $160,000 per year to transfer to the Academy for Research Grants and Publications.

Jeff Hughes, Chair of the Grants Committee, noted that Paul Rothrock was awarded the Welch Award, and no spring applications were received for the Junior Academy grants. Senior Academy members tendered 36 applications requesting $61,924. The Committee granted $31,200 to 24 applications.

Ruth Howes, reporting for the Awards Committee, related that no nominations were made last year. New nomination forms are on the website. Discussion about qualifications for emeritus status resulted in no changes.

Programs and Invitation Chair David Daniell reported that the 2003 Fall meeting will be held at Anderson College around November 1. He called for potential sites for future meetings. He also gave details of the 2002 meeting to be held at Butler University on October 10–11. Abstracts will be due by August 26. All abstracts will be submitted electronically.

Dick Conklin of the Nominations and Elections Committee noted that nominees were need for President-Elect, two for the Grants Committee, and one for the Foundation Committee. Nomination forms are now on the website.

Bill McKnight, head of the Publications Committee, reported that *The Sunflower Family of the Midwest* book is out. Sales are good and he projects $50,000 in sales. *Plants of the Chicago Region* is out of print, with all the original 5000 copies sold. They plan a revised version for 2003. The IAS book inventory is being moved to a safer storage site. He proposed making an agreement with R.G. Balkin

Books to put links to publications on their website to improve individual sales. Book in progress include *Sedges of Indiana*, *Mounds of Indiana*, *Cave Fauna*, and *Geology of Indiana*.

Don Ruch reported for the Biodiversity and Natural Areas Committee that Paul Rothrock was the Winona Welch Award for Botanical Biodiversity Research. The Committee will host two workshops at the Fall meeting. A workshop on Indiana sedges and other graminoids and a workshop about museum techniques for small collections will follow last year's successful darter identification efforts. The Indiana Biological Survey (INDG) website will come online this summer, and an electronic newsletter will be created. A motion to include corporate sponsors in IAS publications was referred to the Publications Committee.

The Junior Academy under Tina Gilliland will hold its annual meeting at Indiana University on November 15. A report about the 2001 meeting was tendered.

Holly Oster of the Library Committee noted that the State Library is moving and IAS items are packed and unavailable. She mailed the revamped *Proceedings* volume and is ready to mail volumes 108, 109, 110, and 111.

Marsha Gillette called for judges for the Science Talent Search to be held October 25–26 at Indiana University Kokomo.

Nelson Shaffer requested approval of a resolution acknowledging the hosts of the Fall Meeting at Butler and one recognizing an Indiana high school finalist in the national science talent search. Both were accepted.

Duvall Jones of the Youth Activities Committee discussed improvement of the high school grant procedure and that for teacher fellowships.

Marsh Moore reported on the latest IAS website improvements.

Damian Schmelz reported for the Natural Resources Commission that John Gross is the new leader of DNR. There is concern about State budgets. The new State Museum will open May 22.

Richard Kjonaas, IAS representative to the AAAS, reported on the annual meeting in Boston where a resolution about cloning was passed. A commercial firm, Ingenta, is seeking to include state academy journals on its website. This possibility was referred to the *Proceedings* editor.

Historian William Gommell called for resumes of current members. He reported the passing of three IAS fellows, 1) James B. Kock of Earlham, 2) Ernest M. Schull of North Manchester, and 3) Harry M. Galloway of Purdue University. A moment of silence in their honor was observed.

New Business

President-Elect Bob Waltz suggested five initiatives. He plans to evaluate the five-year plan to improve Academy operations which was begun under Ruth Howes. He suggested that the time has come for a State-wide Invasive Species Council. He also will seek to better recognize volunteer services of Academy members. Section chairs will be encouraged to show leadership in communications about their disciplines. Finally, improvements in newsletter presentation will be investigated.

President Terry West thanked attendees and adjourned the meeting shortly after 4:00 P.M.

EXECUTIVE COMMITTEE
Butler University
10 October 2002

President Terry West called the meeting to order at 1:04 P.M. Attending: Edward Frazier, Treasurer; Nils I. Johansen, Secretary; Jim Berry, Editor of the *Proceedings*; Frank Guthrie, Foundation Chair; Bob Waltz, President-Elect; Nelson Shaffer, Executive Director; Uwe Hansen, Newsletter editor; Duvall Jones, Bill McKnight, Holly Oster, Jeff Hughes.

Approval of agenda.—Agenda approved. Council meeting not included at this time.

Approval of minutes.—Minutes from Spring meeting at Butler University approved as corrected. Unanimous approval. The Secretary appreciated that the Executive Director had taken notes in the Secretary's absence.

Treasurer's report.—Frazier passed out financial reports. The deficit in operating budget up to $15,000 for a five year period will be covered by funds from the Foundation.

Executive Director's report.—"We are holding our own, i.e., 809 members total." Concern: Corporate members. Nelson Shaffer stressed the need to explore this further. Corporate dues are $250 per year. Some discussion ensued on how to increase membership, but no specific action taken. The current term of the Executive Director is up, and he is willing to continue. Possible renewal will be taken up at the budget meeting in December.

***Proceedings* Editor's report.**—Berry reported that we are again being listed by the major indexing services. Two regular issues for 2002 (will include minutes for 2000 and 2001). Volumes 108/109 will be mailed out soon. Turn-around time for accepted manuscripts is about one-half year.

Newsletter Editor's report.—Main concern is deadlines. Deadlines will be adjusted to fit the IAS calendar.

Secretary's report.—Johansen again thanked Nelson Shaffer for taking minutes at the previous meeting.

President-Elect's report.—Submitted written report. Waltz has met with Nelson Shaffer *re.* program issues. Discussion followed regarding the role and use of Section Chairs. One suggestion was that Section Chairs give information on their sections to the Newsletter Editor. Feedback requested on this.

Another discussion issue was whether IAS should have an institutional representative at each institution and State agency. Feedback requested.

President's report.—West submitted written report. He expressed appreciation for the use of the 'President's Column' in the Newsletter.

Committee Reports

Academy Foundation report.—Guthrie submitted a written report.

Research Grant report.—Hughes reported that 20 grant applications were received, and 13 were approved for a total of $23,489. There are 27 Junior Academy applications pending. It was noted that 24 of these applications came from three high schools.

The total budget for the Junior Academy grants is $5,000. Waltz suggested recognizing Duvall Jones for his work with IAS. The awards committee will send its nomination to the IAS Council. Some discussion followed.

Adjourn.—Meeting adjourned at 3:03 P.M.

Respectfully submitted,
Nils I. Johansen, P.E.
Secretary

COUNCIL MEETING
Butler University
10 October 2002

President Terry West called the meeting to order at 3:21 P.M. Attending: Edward Frazier, Treasurer; Nils Johansen, Secretary; Jim Berry, Editor of the *Proceedings*; Frank Guthrie, Academy Foundation Chair; Bob Waltz, President-Elect; Nelson Shaffer, Executive Director; Duvall Jones, Youth Activities Chair; Bill McKnight, Publications Committee Chair; Holly Oster, Librarian; Uwe Hansen, Newsletter Editor; Jeff Hughes, Research Grants Chair; Marcia Moore, Web Site Coordinator; Rebecca Dolan, Past-President; John Schutt, Speaker of the Year Committee; Dave Daniell, Local Arrangements and Programs Chair; Stan Shimer, Botany Section Chair; Blake Janutolo; Don Ruch, BNC Chair; Dick Conklin, Nominations and Elections Chair; William Gommel, Historian; Richard Kjonaas, AAAS Representative.

Approval of minutes.—Minutes from the Spring Meeting at Butler University approved as corrected. Unanimous approval. The Secretary expressed appreciation to the Executive Director for taking minutes in the Secretary's absence.

Approval of agenda.—Agenda approved as presented.

Treasurer's report.—Frazier passed out financial reports. The deficit in the operating budget is to be covered by up to $15,000 per year for a five year period from the Foundation funds.

Executive Director's report.—"We are holding our own, i.e., 809 members total." Concern: Corporate members. Shaffer stressed the need to explore this further. Corporate dues are $250 per year. Some discussion ensued on how to increase membership, but no specific action taken. Shaffer noted that the current term of the Executive Director is up, and he is willing to continue in office. Possible renewal will be taken up at the budget meeting in December.

***Proceedings* Editor's report.**—Berry reported that the *Proceedings* is again being listed by the major indexing services. There will be two regular issues for 2002, and will include the Academy minutes for 2000 and 2001. Volumes 108/109 will be mailed out soon. Turnaround time for manuscripts in the *Proceedings* is about ½ year.

Newsletter Editor's report.—The main concern is deadlines. Deadlines will be adjusted to fit the IAS calendar.

Secretary's report.—Johansen noted that Nelson Shaffer had taken minutes at the previous meeting.

President-Elect's report.—Submitted written report. Waltz has met with Nelson Shaffer regarding program issues. Discussion followed regarding the role and use of Section Chairs. One suggestion was that Section Chairs give information on their sections to the Newsletter Editor. Feedback requested on this. Another discussion issue: Should the IAS have an institutional representative at each institution and State agency? Feedback is requested.

The Annual Budget Meeting will be 9 November at Butler University at 10 AM.

Conklin moved, Berry seconded, that Duvall Jones receive a Special Service award. Approved.

President's report.—West submitted a written report. He expressed appreciation for the use of the 'President's Column' in the Newsletter.

Committee Reports

Academy Foundation report.—Guthrie submitted a written report.

Amendments.—Hansen suggested that the process for election to Emeritus Member be simplified to: Age 65, retired, 25 or more years of membership. These points, when verified by the Executive Director, would automatically make a member an Emeritus Member if he or she so desired. The vote/comments would be taken up at the Spring Meeting 2003.

Awards.—Shaffer read the report from Howes: Distinguished Scholar: Harry G. Day; Fellow: James Bandoli, Christian Johannsen, Shrikrishna Dhawale; Emeritus: Chris Ault, Winifred Caponigri, Jon Hendrix, Uwe Hansen, Benjamin Swartz. Unanimous approval.

Research Grants.—Hughes reported that 20 grant applications were received, and 13 were approved for a total of $23,489. There are 27 Junior applications pending. Note that 24 of these applications came from three high schools. The total

budget for the Junior Academy grants is $5000. Waltz suggested recognizing Duvall Jones for his work for the Junior Academy of the IAS. The Awards Committee will send his nomination to the IAS Council. Some discussion followed.

Programs and invitations.—The Spring and Fall meetings in 2003 will be at Anderson University. The dates are 4-5 April and 16-17 October 2003.

Nominations and elections.—The election results are: Frazier – Treasurer; Guthrie – Foundation; Don Ruch – President-Elect; David Hicks – Research grants. Next year's openings: President-Elect, Secretary, Research Grants, Foundation.

Publications.—McKnight summarized the current activities. He was asked to look into corporate sponsorship and page charges and report back at the next meeting.

Finance.—IRS audit was passed.

Resolution.—A resolution thanking Butler University for hosting the meeting was read and approved for presentation at the general membership meeting.

Youth activities.—There was a written report from Jones.

Library.—Oster reported on the status of the State Library and IAS' place therein.

Biodiversity.—Members were reminded about the special sessions and workshop at the Fall Meeting.

Web site.—It is up and running. The Council instructed Executive Director to poll the members *re.* inclusion in an electronic membership directory. (Nelson moved, Ruch seconded, approved, with one nay.)

Speaker of the Year.—Schutt needs names of potential candidates for Speaker of the Year for 2003.

AAAS report.—Kjonaas will attend the AAAS meeting in February 2003.

Historian.—Gommel read the names of members who have passed away over the last year.

New Business

The Council directed the Executive Director to look into guidelines regarding corporate members. The budget for this should not exceed $1000. (Nelson moved, Johansen seconded, approved).

The Council thanked Duvall Jones for his service to the Youth Activities Committee.

The meeting adjourned at 6:02 PM.

Respectfully submitted,
Nils I. Johansen, P.E.
Secretary

INDIANA ACADEMY OF SCIENCE

2002 Year End Financial Report

	Balance 1-Jan-02	Revenues	Expenses	Balance 31-Dec-02
OPERATING FUND	6,050.46			7,851.37
Dues		18,000.00		
Interest		1,227.13		
Contributions		215.00		
Spring Meeting		375.00	373.00	
Annual Meeting		6,651.00	7,921.67	
Transfer from IAS Foundation		9,000.00		
Misc. Income		730.00		
Officer's Expenses			4,000.00	
Operating Expenses			6,278.82	
Financial Expenses			1,026.00	
Newsletter Expenses			3,876.62	
Library Operations			0.00	
Committee Expenses			0.00	
Speaker of the Year			835.00	
AAAS Representative			875.00	
Web Site Development			1,000.00	
Youth Activities			8,211.11	
Operating Fund Total	6,050.46	36,198.13	34,397.22	7,851.37
RESTRICTED FUNDS				
Proceedings	38,830.08	17,421.77	38,101.14	18,150.71
Publications	(804.06)	75,866.68	61,655.19	13,407.43
* Research Grants	8,704.33	68,706.09	63,207.45	14,202.97
Indiana Biological Survey	0.00	2,000.00	2,000.00	0.00
Lilly Library	6,578.44	0.00	0.00	6,578.44
Welch Fund	9,906.01	0.00	400.00	9,506.01
Life Members Fund	3,795.25	500.00	0.00	4,295.25
Past Presidents Fund	9,178.39	0.00	0.00	9,178.39
Total Restricted Funds	76,188.44	164,494.54	165,363.78	75,319.20
Prepaid 2003 Dues				4,255.00
TOTAL FUNDS	**82,238.90**	**200,692.67**	**199,761.00**	**87,425.57**
FUNDS ON DEPOSIT				
Checking Account	14,262.86	191,460.59	200,398.00	5,325.45
Money Market Savings	57,854.32	100,209.42	87,020.00	71,043.74
Cert. of Deposit	10,121.72	934.66	0.00	11,056.38
TOTAL FUNDS DEPOSITED	**82,238.90**	**292,604.67**	**287,418.00**	**87,425.57**

* Provided 36 senior member grants, 16 high school grants and 2 high school teacher fellowship

ACADEMY FOUNDATION FUNDS				
John S. Wright Fund Account	6,258,529.00			5,990,130.00
Academy Fund Account	207,840.00			225,284.00
Invested Income Account	176,527.00			230,011.00
TOTAL FOUNDATION FUNDS	**6,642,896.00**			**6,445,425.00**

Foundation Funded Used For:

General Fund	9,000.00	
Proceedings	5,912.56	
Publications	42,555.03	Edward L. Frazier
Research Grants	66,271.80	Treasurer
Total	123,739.39	

INSTRUCTIONS TO AUTHORS

(revised October 2001)

General information.—Manuscripts and all correspondence should be sent to the Editor. To be eligible for publication in the *Proceedings*, at least one author must be a member of the Academy. Papers that have been presented at an Academy meeting may be given preference for publication. Submission of a manuscript to the Editor implies that it has not been sent to another publication, nor has it been published elsewhere. If it is accepted for publication in the *Proceedings*, it may not be republished without the written consent of the Academy.

All manuscripts are evaluated by referees. Submit three copies of the manuscript. Figures in manuscripts for review may be submitted as photocopies, and they should be approximately the size that they will be printed in the *Proceedings*.

After the manuscript has been accepted, the author will be asked to submit the manuscript as a paper copy and on a computer disc in a widely-used word processing program. Indicate clearly on the computer disc the **word processing program** and the **type of computer** (Mac or PC).

Galley proofs will be sent to the primary author for approval and correction.

Style.—Do not use right or full justification. Also, use a non-proportional font (e.g, Courier, but not Times Roman).

Title page.—The title page should include (1) the title in capital letters, (2) each author's name and address, (3) the running head (see below), and (4) the complete name, address, and telephone number of the author with whom proofs and correspondence should be exchanged, a FAX number and electronic mail address if available.

Abstract.—All manuscripts have an abstract, which should summarize the significant facts in the manuscript. The "ABSTRACT" heading in capital letters should be placed at the beginning of the first paragraph set off by a period. Use complete sentences, and limit the abstract to one paragraph and 250 words.

Keywords.—Give 3–5 appropriate keywords following the abstract.

Text.—Double-space text, tables, legends, etc. throughout. Three categories of headings are used. The first category (METHODS, RESULTS, etc.) is typed in capitals, centered, and on a separate line. The second (lower) category of heading, in bold type, begins a paragraph with an indent and is separated from the text by a period and a dash.(This paragraph begins with an example of this heading.) The third heading category may or may not begin a paragraph, is italicized and followed by a colon. (The paragraph below is an example.) Use only the metric system unless quoting text or referencing collection data.

Citation of references in the text: Cite only papers already published or in press. Include within parentheses the surname of the author followed by the date of publication. A comma separates multiple citations by the same author(s) and a semicolon separates citations by different authors, e.g., (Smith 1990), (Jones 1988; Smith 1993), (Smith 1986, 1987; Smith & Jones 1989; Jones et al. 1990).

Literature cited section.—Use the following style, and include the full unabbreviated journal title. Repeat the name for multiple references by the same author. Note that book titles have the first letter of each word capitalized.

Walter, J. & B. Hallet. 1979. Geometry of former subglacial water channels and cavities. Journal of Glaciology 23:335–346.

Walter, J. 1992. The significance and complexity of communication in moths. Pp. 25–66, *In* Insect Communications: Mechanisms and Ecological Significance. (P.N. Work & J.S. Rivers, eds.). Princeton University Press, Princeton, New Jersey.

Footnotes.—Footnotes are permitted only on the first printed page to indicate current address or other information concerning the author. These are placed together on a separate page at the end of the manuscript. Tables and figures may not have footnotes.

Running head.—The author's surname(s) and an abbreviated title should be typed all in capital letters and must not exceed 60 characters and spaces. The running head should be placed near the top of the title page.

Tables.—These should be typed double-spaced, one table to a page and numbered consecutively. Most tables contain only three horizontal lines (see recent issues for examples). Do not use vertical lines or shading. Include all pertinent information in the table legend (no footnotes).

Illustrations.—All art work must be camera-ready (mounted and labeled) for reproduction. Figures should be arranged so that they fit (vertically and horizontally) the printed journal page, either one column or two columns, with a minimum of wasted space. When reductions are to be made by the printer, pay particular attention to width of lines and size of lettering in line drawings. Multiple photos assembled into a single plate should be mounted with only a minimum of space separating them. In the case of multiple illustrations mounted together, each illustration must be numbered Fig. 1, Fig. 2, etc.) rather than given letter (A,B, etc.) designations. The name(s) of author(s), and an indication of top edge, and whether the illustration should be one or two columns wide, should be written on the back of the illustration. The overall dimensions should be no more than 11 inches (28 cm) × 14 inches (36 cm). Larger drawings present greater difficulty in shipping and greater risk of damage for which the *Proceedings* assumes no responsibility. In manuscripts for review, photocopies are acceptable, and should be reduced to the exact measurements that the author prefers in the final publication. Make notations in the text margins to indicate the preferred position of illustrations in the printed text. Color plates can be printed; but the author must assume the full cost, currently about $600 per color plate.

Legends for illustrations should be placed together on the same page(s) and separate from the illustrations. Each plate must have only one legend, as indicated below:

Figures 1–4.—Right chelicerae of species of *Centruroides* from Timbuktu. 1. Dorsal view; 2. Prolateral view of moveable finger; 3. *Centruroides* holotype male; 4. *Centruroides* male. Scale = 1.0 mm.

Page charges and reprints.—Page charges are currently $10 per journal page for members of the Academy and $70 per page for non-members. Corrections in proof pages must be restricted to printer's errors only; all other alterations will be charged to the author (currently $3 per line).

Reprints are available only from the Allen Press and should be ordered (with payment) when the author receives the proof pages. Allen Press will not accept reprint orders after the paper is published.

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